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ORIGINAL ARTICLES

PROPERTIES OF SUB-FRACTIONS OF HYDROGEN CLAY PREPARED FROM INDIAN SOILS, I*

BY

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(Received for publication on 25 August 1941)

(With six text-figures)

IN recent years, the properties of sub-fractions of clay having particles of different sizes have attracted considerable attention in consequence of the light they throw on soil formation processes [Brown and Byers, 1932; Bradfield, 1935; Bray, 1937]. Apart from this standpoint, variations in colloidal properties with the particle size, even when the mass chemical composition of the disperse phase, the clay, shows little or no change with the degree of dispersion, are of considerable theoretical interest. A study of the properties of sub-fractions of hydrogen clays separated from Indian soils has been undertaken by us with both these objects in view. The present paper is the first of a series which will deal with these studies.

EXPERIMENTAL

(a) *Separation of sub-fractions from the entire clay fraction*—Particulars regarding the soils used are given below.

TABLE I
Particulars of soils used

Laboratory No.	Description of soil	pH		Clay + loss on solution (per cent)	Silt (per cent)	Base-exchange capacity by Parker's [1929] method	Total exchangeable bases (S) by Rice Williams' [1932] method
		Aq. suspension	NKCl suspension				
22	Red lateritic soil from Government Agric. Farm, Dacca (Bengal) collected from a depth of 0 to 6 inches	5.12	4.21	22.8	30.1	8.4	4.8
34	Black soil from Government Experimental Farm, Akola, Berar (C. P.) collected from a depth of 0 to 9 inches	7.10	7.05	44.8	19.6	39.5	39.5

*The results given in this paper have been taken from the published Annual Report for 1937-38 on the working of a scheme of research into the Properties of Colloid Soil Constituents financed by the Imperial Council of Agricultural Research, India

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From the entire clay fractions of the soils the following subfractions were separated by controlled centrifugal subsidence. A 'bucket type' centrifuge* having a diameter of 25 cm. and capable of making 5,000 revolutions per minute was used.

TABLE II

Equivalent spherical diameters of sub-fractions and their percentages in the entire clay fraction

Soil	Reference number of sub-fraction	Percentage of sub-fraction in the entire clay	Limiting equivalent spherical diameters in microns	Reference number of corresponding hydrogen clay
1. Red lateritic soil	1	25.10	0.26 and 2.0	L ₁
	2	9.60	0.16 and 0.26	L ₂
	3	65.40	< 0.16	L ₃
2. Black soil	1	13.50	0.45 and 2.0	M ₁
	2	4.80	0.23 and .045	M ₂
	3	5.20	0.15 and 0.23	M ₃
	4	75.50	< 0.15	M ₄

The depth of sampling and the rate of revolution of the centrifuge were kept constant in separating the different fractions; only, the time of settling was varied. The limiting equivalent spherical diameters (given in column 4 of Table II) were calculated from Stokes' law under the following simplifying assumptions:

(i) The different fractions have the same density.

(ii) The particles settle under a uniform centrifugal force, viz. that obtaining at half height.

(iii) The particles have a spherical symmetry.

The different subfractions were leached with dilute (0.05N) hydrochloric acid to obtain the corresponding hydrogen clays which were washed free from HCl and made up to suspensions containing 2.5 gm. of oven-dried (105°C.) material per litre. All measurements reported in this paper were made with these hydrogen clays.

(b) *Fusion analysis for Fe, Al and Si.*—The usual methods of soil and clay analysis were followed [Wright, 1937].

(c) *Electrometric titration of hydrogen clays.*—The technique of Mukherjee *et al.* [1936] was followed. In addition to hydrogen and quinhydrone electrodes used by them glass electrodes (Morton type) in conjunction with a Cambridge electrometer valve potentiometer reading directly to 2 millivolts were used.

*A Sharples supercentrifuge is now being used for separating very fine sub-fractions

RESULTS

(a) *Chemical composition.*—The results of fusion analysis are given in Table III.

TABLE III

Chemical compositions of the entire hydrogen clay and its sub-fractions isolated from the Dacca soil

Hydrogen clay	SiO ₂ per cent	Al ₂ O ₃ per cent	Fe ₂ O ₃ per cent	SiO ₂	SiO ₂
				Al ₂ O ₃	Al ₂ O ₃ + Fe ₂ O ₃
L *	51.2	36.0	11.9	2.40	1.99
L ₁	64.9	30.6	5.0	3.60	3.26
L ₂	49.6	36.5	12.6	2.31	1.85
L ₃	39.9	46.5	14.0	1.46	1.22

*Obtained from the entire clay fraction

TABLE IV

Chemical compositions of the entire hydrogen clay and its sub-fractions isolated from the Akola soil

Hydrogen clay	SiO ₂ per cent	Al ₂ O ₃ per cent	Fe ₂ O ₃ per cent	SiO ₂	SiO ₂
				Al ₂ O ₃	Al ₂ O ₃ + Fe ₂ O ₃
M*	53.1	30.4	16.9	3.0	2.19
M ₁	65.8	24.2	9.8	4.6	3.7
M ₂	59.3	26.9	14.2	3.7	2.6
M ₃	52.1	30.6	18.0	2.9	2.1
M ₄	48.4	32.3	19.3	2.6	1.8

*Obtained from the entire clay fraction

The percentage of silica decreases with diminishing particle size while that of alumina and ferric oxide increases. In consequence, the silica-alumina and silica-sesquioxide ratios decrease with diminishing particle size. The percentage of silica is definitely higher in the coarsest fraction of the red soil and the two coarse fractions of the black soil compared with the respective entire clays. The percentages of Al₂O₃ and Fe₂O₃ are, on the other hand, greater in the entire clays than in the above coarse fractions but lower than in the finer fractions.

The results of fusion analysis of the silt fractions given in Table V follow the regular sequence of variations in composition with the particle size within the clay fraction.

TABLE V

Chemical compositions of the silt of the Dacca and Akola soils

	SiO ₂ (per cent)	Al ₂ O ₃ (per cent)	Fe ₂ O ₃ (per cent)
Silt of the Dacca soil	90.4	6.76	1.42
Silt of the Akola soil	80.2	12.40	6.02

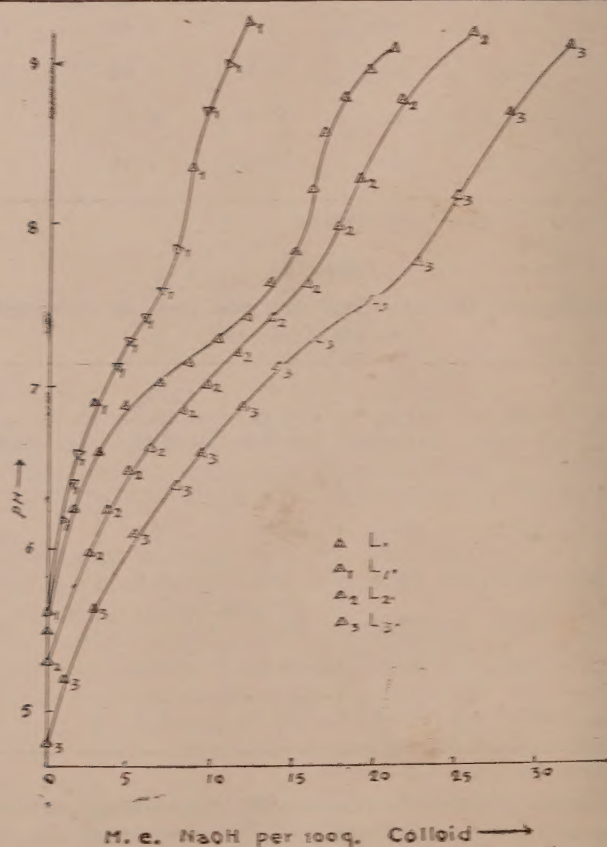


FIG. 1. Titration curves with NaOH of the entire hydrogen clay and its sub-fractions isolated from the Dacca lateritic soil

A similar increase in the percentage of silica in the silt fraction as compared with the 1-2 μ fraction was observed by Marshall [1935]. He attributed this to a greater percentage of free quartz in the silt.

The decrease in the percentage of silica with diminishing particle size indicates that the chemical weathering process attending the mechanical break-down of the large particles is one of continued desilication which appears to be more pronounced with the red lateritic soil than the black soil in agreement with the more intense conditions of leaching under which the red soil has been formed compared with the other soil.

(b) *Free and total acids.*—Table VI gives the free acids calculated from the pH's of 0.25 per cent suspensions and the total neutralizable acids calculated from the titration curves with different bases given in Figs. 1-6. The total acids have been calculated both at the inflexion points of the curves as also at pH 7.0.

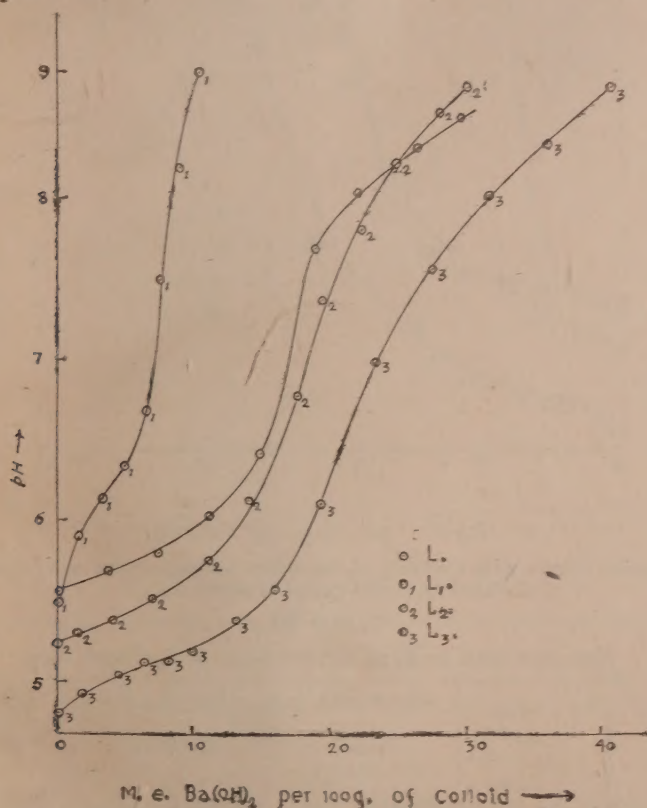


Fig. 2. Titration curves with Ba (OH)₂ of the entire hydrogen clay and its sub-fractions isolated from the Dacca lateritic soil

Both the free and total acids increase with diminishing particle size. L₁ and M₁ obtained from the coarsest fractions have much smaller total acids than L and M obtained from the entire clays. L₂ and L₃ have greater total acids than L. Those of M₂ and M₃ are small compared with M. M₄, however, gives a definitely greater value than M. The larger total acid of M compared with M₁, M₂ and M₃ is expected as the entire clay fraction is largely made up of the finest fraction (Table II) having the largest total acid.

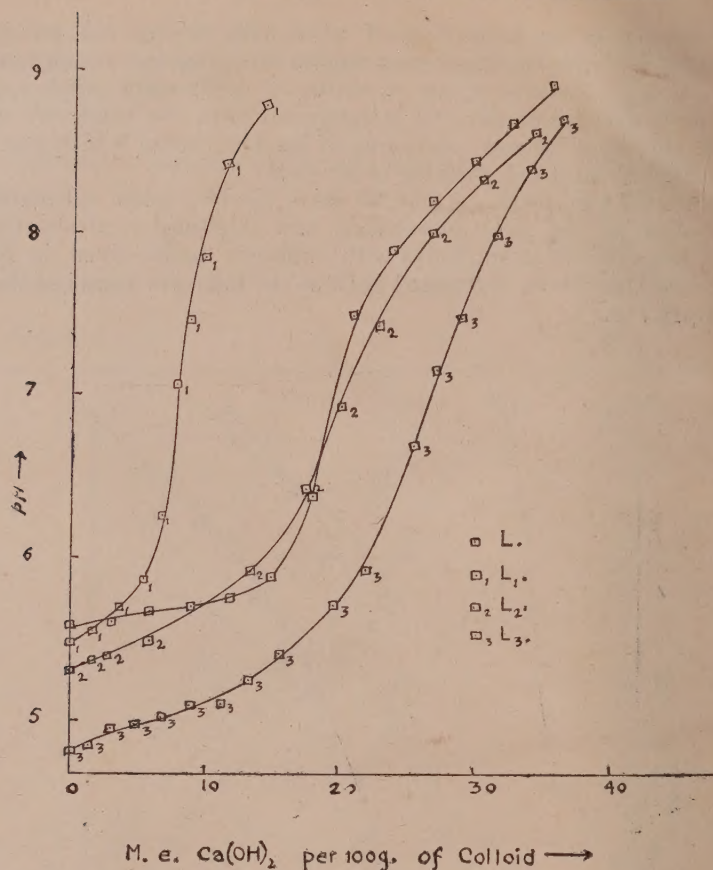


FIG. 3. Titration curves with $\text{Ca}(\text{OH})_2$ of the entire hydrogen clay and its sub-fraction isolated from the Dacca lateritic soil

TABLE VI

Free and total acids of sub-fractions of hydrogen clay

Hydrogen clay	$\frac{\text{SiO}_2}{\text{Al}_2\text{O}_3}$ (molar)	Free acid in m. e. H^+ ions per 100 gm. colloid	Total acid in m. e. base per 100 gm. colloid using					
			NaOH		$\text{Ba}(\text{OH})_2$		$\text{Ca}(\text{OH})_2$	
			At inflexion point	At pH 7.0	At inflexion point	At pH 7.0	At inflexion point	At pH 7.0
L_1	3.60	0.07	8.33	3.30	7.0	7.0	8.0	8.0
L_2	2.31	0.17	17.60	9.03	18.0	18.6	20.2	20.2
L_3	1.46	0.60	24.00	12.65	19.6	23.8	26.0	26.6
M_1	4.60	0.08	3.80	3.70	2.8	5.0	3.5	5.3
M_2	3.70	0.21	14.00	10.00	12.6	13.0	13.6	14.7
M_3	2.9	1.10	36.5	31.50	30.5	33.5	31.5	38.5
M_4	2.5	4.38	86.0	86.00	86.0	97.0	90.0	98.5

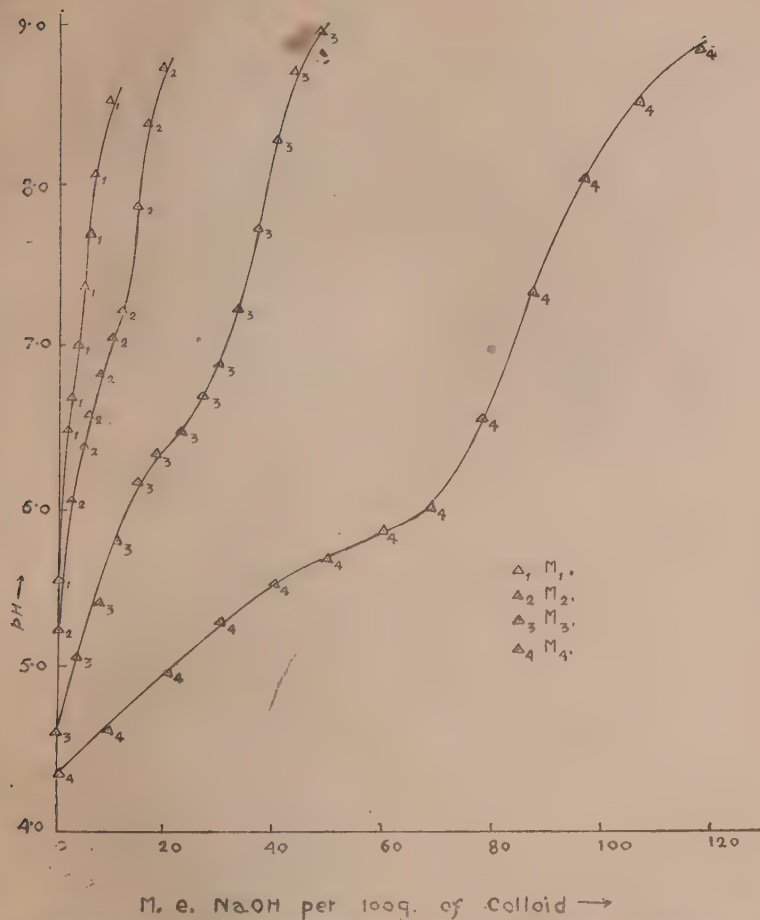


FIG. 4. Titration curves with NaOH of sub-fractions of hydrogen clay isolated from the Akola soil

In Table VII the observed total acids of L and M have been compared with their total acids calculated from those of the sub-fractions and the percentages of these sub-fractions in the entire clays.

TABLE VII

Total acids of entire hydrogen clays calculated from their titration curves and from those of their sub-fractions

Hydrogen clay	Total acid (at pH 7.0) in m. e. base per 100 gm. colloid using					
	NaOH		Ba(OH) ₂		Ca(OH) ₂	
	Observed	Calculated	Observed	Calculated	Observed	Calculated
L	6.3	9.9	17.0	19.0	19.5	21.3
M	60.0	67.0	79.0	75.2	87.0	77.0

The observed and calculated values show a fair agreement ignoring the total acids of L with NaOH and of M with $\text{Ca}(\text{OH})_2$.

(c) *Form of titration curves.*—Previous work from this laboratory has dealt with the titration curves of hydrogen clays obtained from entire clay fractions [Mitra, 1936, 1940; Mukherjee, Mitra and Mukherjee, 1937]. A comparison of the titration curves of sub-fractions of hydrogen clay obtained from the same soil may afford useful information regarding the nature of the reactive acidic material present in the different fractions. Such comparative studies have not been previously made.

The NaOH curves of L_1 , L_2 and L_3 given in Fig. 1 are similar to one another and to that of L obtained from the entire clay fraction. This is also true of M_1 , M_2 , M_3 , M_4 and M given in Fig. 4. This similarity suggests that the same acid is being titrated in the different sub-fractions. This conclusion, however, does not apparently harmonize (further discussed later) with their markedly different chemical compositions given in Tables III and IV.

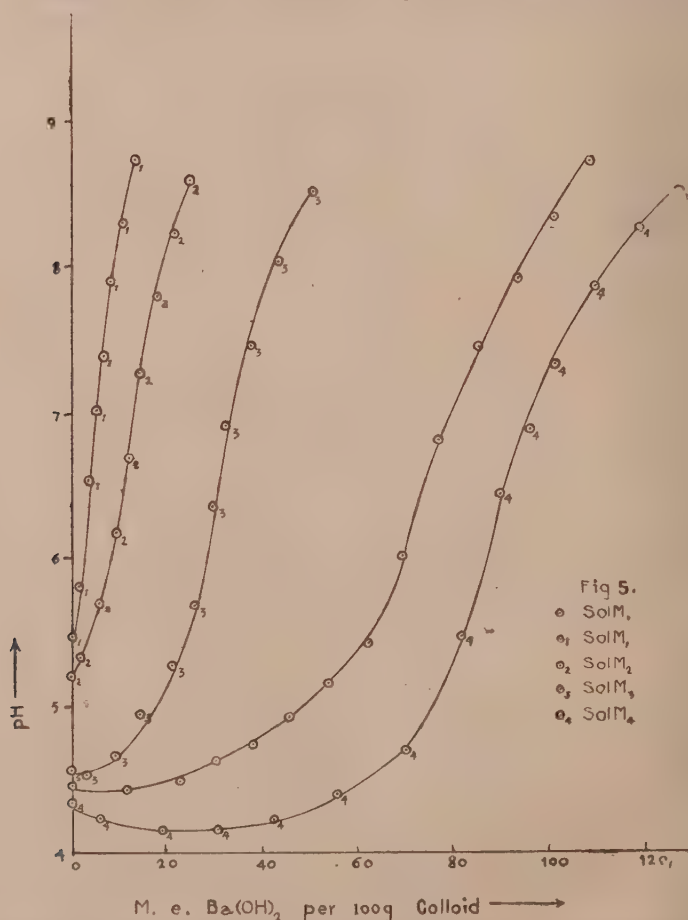


FIG. 5. Titration curves with $\text{Ba}(\text{OH})_2$ of the entire hydrogen clay and its sub-fractions isolated from the Akola soil

The NaOH curves all have an apparent weak monobasic acid character. Table VIII shows the pH at their inflexion points and the apparent dissociation constants calculated from the pH 's at half neutralization.

TABLE VIII

pH at inflexion and dissociation constants of hydrogen clays

Sol	pH at inflexion	$pH (= pK)$ at half neutralization	K
L	8.21	7.13	7.41×10^{-8}
L ₁	8.10	7.15	7.10×10^{-8}
L ₂	8.00	6.97	1.07×10^{-7}
L ₃	8.03	6.96	1.10×10^{-7}
M	7.49	6.20	6.31×10^{-7}
M ₁	7.10	6.56	2.75×10^{-7}
M ₂	7.63	6.66	2.20×10^{-7}
M ₃	7.60	6.26	5.50×10^{-7}
M ₄	7.00	5.56	2.75×10^{-6}

The inflexion points in the titration curves of L, L₁, L₂ and L₃ occur within narrow range of pH (8.00-8.21) and the dissociation constants calculated from the curves are in fair agreement. With M, M₁, M₂, M₃ and M₄ the inflexion points are located within a wider range of pH (7.00-7.63) though the agreement between the dissociation constants is not unsatisfactory excepting the finest fraction (M₄) whose dissociation constant is ten times that of the coarsest. The dissociation constant has a tendency to increase with diminishing particle size. This is true of the sub-fractions obtained from the soil.

The buffer capacity curves of the various sub-fractions obtained on plotting the buffer capacity, β ($\beta = \Delta\beta/\Delta pH$) against the amount of the base (NaOH) added show definite maxima. In Table IX the maximum buffer capacities (β max.) of the various sub-fractions, as also the pH and the percentage neutralization at maximum buffer capacity, have been compared.

TABLE IX

Maximum buffer capacity, and pH and per cent neutralization at maximum buffer capacity of hydrogen clays

Sol	β max.	pH at β max.	Per cent neutralization at β max.
L ₁	5.8	7.45	69.6
L ₂	11.0	7.20	62.5
L ₃	19.0	7.50	79.1
L	10.5	7.30	64.6
M ₁	5.0	6.68	65.7
M ₂	10.5	6.90	71.4
M ₃	26.0	6.70	71.2
M ₄	66.0	5.63	60.5
M	50.0	6.45	66.0

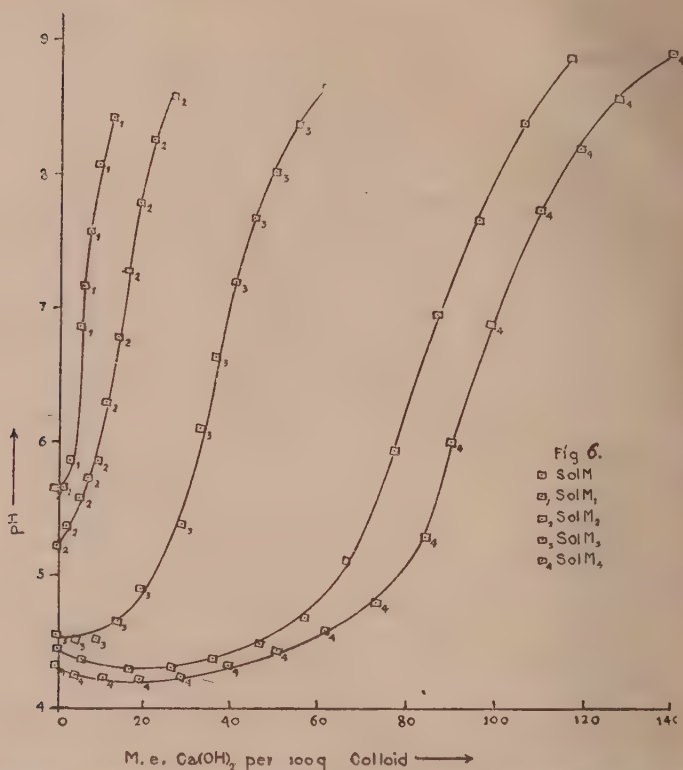


Fig. 6. Titration curves with $\text{Ca}(\text{OH})_2$ of the entire hydrogen clay and its fractions isolated from the Akola soil

The finer the fraction, the higher is β max. With L_1 , L_2 , L_3 and L , β max. occurs within a small range of pH . This is also true of M_1 , M_2 , M_3 and M . β max. of M_4 , however, occurs at a much lower pH compared with the other hydrogen clays. It is to be noted that β max. in no case corresponds to 50 per cent neutralization as would be expected in the case of a dissolved weak acid. It occurs at a higher stage of neutralization (60 to 80 per cent). Evidently, the interaction of the hydrogen clays with the base is not simple neutralization of H^+ by OH^- ions. It is complicated by other factors [Mukherjee, Mitra and Mukherjee, 1937.]

The $\text{Ba}(\text{OH})_2$ curves of L_1 , L_2 , L_3 and L given in Fig. 2 are not all quite similar. Those of L_1 and L_3 have an apparent weak acid character. L and L_2 on the other hand, behave as strong acids. The dissociation constant calculated from the pH at half neutralization is 6.7×10^{-7} for L_1 and 5.5×10^{-6} for L_3 . The finer fraction thus behaves as a stronger acid.

The $\text{Ca}(\text{OH})_2$ curves of L , L_1 and L_2 have the same form which is different from that of sol L_3 (Fig. 3).

The $\text{Ba}(\text{OH})_2$ and $\text{Ca}(\text{OH})_2$ curves of M_1 , M_2 , M_3 , M_4 and M given in Figs. 5 and 6 show a general similarity of form. Those of M_1 , M_2 , M_3 and M_4 present one peculiar feature, viz. that on the first addition of the base, the

H of the sol instead of increasing or remaining constant shows a definite decrease. This is specially marked with the baryta curves of the finer fractions. Acid appears to be liberated as a result of the interaction between the sol and the base, a result which is foreign to classical principles of acid-base interactions. An explanation of the observation is deferred pending completion of further investigations now in progress.

(d) *Total acidity per gramme and per sq. cm. of surface.*—The increase in the total acid with diminishing particle size (Table VI) may be referred to an increase in the specific surface and hence to a greater number of exchange pots in a given area. In Table X the total acids of the sub-fractions from the black soil calculated per gramme (T_g) and per sq. cm. of the surface (T_s) have been compared.

TABLE X

Specific surface and b. e. c. per gm. and per sq. cm. of surface of hydrogen clays

Hydrogen clay	Average sp. surface in sq. cm $\times 10^3$	Total acid in m. e. NaOH at inflexion point	
		per gm. (T_g)	per sq. cm $\times 10^7$ (T_s)
M_1	18.5	0.038	20.5
M_2	66.5	0.140	21.0
M_3	120.0	0.365	30.0
M_4	>160.0	0.860	<55.0

It will be seen that T_s does not show any marked or regular variation with the particle size* though T_g rapidly increases as the latter decreases.

GENERAL DISCUSSION

The variations in the total acid per gramme with the particle size may arise from variations in the nature of the reactive material making up the various fractions as the differences in chemical composition would also suggest. The similarity in the form of the titration curves of the different fractions, especially the NaOH curves, and the fair agreement between the values of the dissociation constant calculated from them, on the other hand, indicate that the active acidic material present in the different fractions is essentially the same. Considered in this light the variations in the total acid (per gramme) and chemical composition may be due to varying admixtures of 'free' silica and sesquioxides having negligible base-combining capacity in the different fractions. The differences in composition might also arise from isomorphous replacements [Marshall, 1935] within the lattice of the constituent minerals and/or differences in relative proportions of several types of

*Variations in T_s have been observed using sub-fractions from other soils. These will be discussed in the next paper of this series.

clay minerals in the different fractions but these would probably give rise to more marked variations in the form of the titration curves than observed in this work.

SUMMARY

The variations in chemical composition, form of titration curves with bases and the base exchange capacities (b.e.c.) calculated from these curves of hydrogen clays prepared from three and four sub-fractions respectively of the entire clay fraction of a red lateritic soil from Dacca (Bengal) and a black soil from Akola (Central Provinces) have been studied. As the particle size decreases, the percentage of Al_2O_3 increases but that of SiO_2 and Fe_2O_3 diminishes as also the silica-sesquioxide ratio. The b. e. c. and the amount of free H^+ ions calculated per gramme rapidly increase with diminishing particle size but calculated per square centimetre of the external surface the b. e. c. shows no marked or regular variation. The different sub-fractions prepared from the same soil give nearly the same type of titration curves.

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SEPARATIONS IN THE PROPERTIES OF HYDROGEN CLAYS ON THE REMOVAL OF FREE INORGANIC OXIDES CONTAINED IN THEM, I *

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(With five text-figures)

Inorganic colloidal material of soil, besides containing Si, Al and Fe in a combined form making up its mineral constituents proper, is often associated with varying amounts of free oxides of these elements. The effect of these free oxides in determining the base exchange and other properties of the soil colloidal material is not very well understood. A systematic study has been undertaken of the changes in properties of hydrogen clays consequent on the removal of the free oxides contained in them. The methods usually used for this purpose are not free from the criticism that they may decompose, or, alter the properties of the clay minerals and may not effect a complete separation of the oxides. In spite of these limitations it is of interest to examine the changes brought about by such separations. The present paper deals with changes consequent on treatments according to the methods of Tamm [1922], Mattson [1931], and Truog and Truog [1935] for the removal of the free oxides. The following properties have been studied : (i) chemical composition, (ii) the form of potentiometric titration curves with bases and (iii) the base exchange capacity calculated from these curves. The hydrogen clays used are listed on the next page. Further results will be reported in subsequent papers of this series.

EXPERIMENTAL

(i) *Separation of the free inorganic oxides.*—Tamm uses a solution of ammonium oxalate having a pH 2.32. In Mattson's method which is used for the separation of the free sesquioxides only the clay is treated with a saturated solution of aluminium chloride. In the method of Drosdoff and Truog, free silica and alumina are first removed by digestion at 70°C., with a 10% solution of sodium carbonate. The free ferric oxide is then removed with a solution of sodium sulphide which is formed on passing H_2S gas through an aqueous suspension of the clay.

The results given in this paper have been taken from the published Annual Reports for 1935-36, 1936-37 and 1938-39 on the working of a scheme of research financed by the Central Council of Agricultural Research, India.

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Reference No. of hydrogen clay (before removal of free oxides)	SiO ₂ /R ₂ O ₃ ratio of hydrogen clay	Lab. No. and description of soil from which hydrogen clay was obtained
F	1.94	High land acid soil from Government Farm, Burdwan (Bengal) collected at a depth of 0-6 inch from Block B, Plot No. 40. Lab. No. 14
I	2.50	Neutral calcareous soil from Satara district (Bombay) collected at a depth of 0-6 inch. Lab. No. 25
K	2.54	Neutral black soil from Bilaspur near Raipur (C. P.) collected at a depth of 0-6 inches. Lab. No. 3
L	1.99	Red lateritic soil from Government Farm at Dacca (Bengal) collected at a depth of 0-6 inches. Lab. No. 22
N	1.88	Bhata red laterite soil from C. P. collected at a depth of 0-9 inches. Lab. No. 33

(b) *Preparation of hydrogen clays.*—Hydrogen clays were prepared from the entire clay fractions (before and after removal of their free oxides) by leaching them with 0.02N hydrochloric acid. In the sequel, the subscripts *b* and *c* to the reference numbers of hydrogen clays have been used to denote those prepared after removal of the free oxides by the methods of Mattson, Tamm, and Drosdoff and Truog respectively.

(c) *Chemical analysis.*—The percentages of SiO₂, Al₂O₃ and Fe₂O₃ in the hydrogen clays have been determined after fusion with sodium carbonate [Wright, 1937].

(d) *Electrometric titration of hydrogen clays.*—Details of procedure adopted for this work have been described elsewhere [Mukherjee *et al.*, 1936 ; Mitra, 1936, 1940]. Hydrogen and glass electrodes were used.

RESULTS

A. Hydrogen clays prepared from the Burdwan Farm soil (Lab. No. 14)

The free oxides were removed by Tamm's method. Figs. 1 and 2 give the titration curves with bases. The base exchange capacities* (b.e.c.) calculated from the curves are given in Table I and the results of fusion analysis in Table II.

TABLE I

Base exchange capacities of hydrogen clay from Burdwan soil before and after removal of free oxides

Hydrogen clay	Base used for titration	pH at inflexion	B.e.c. in m.e. base per 100 gm. colloid at inflexion point of titration curve
F	NaOH	6.15 ; 7.85	9.0 ; 31.6
	Ba (OH) ₂	7.0	31.0
F ^b	NaOH	4.7 ; 7.2	3.0 ; 25.0
	Ba (OH) ₂	5.0 ; 6.2	7.5 ; 25.0
	Ca (OH) ₂	6.2	25.0

* Reproducible to within ± 2.5 per cent

TABLE II

Chemical composition of hydrogen clay from Burdwan soil before and after removal of free oxides

hydrogen clay	SiO ₂ per cent	Al ₂ O ₃ per cent	Fe ₂ O ₃ per cent	SiO ₂ /Al ₂ O ₃ (molar)	SiO ₂ /R ₂ O ₃ (molar)
. . .	49.2	29.2	21.4	2.85	1.94
. . .	51.5	30.9	17.4	2.82	2.01

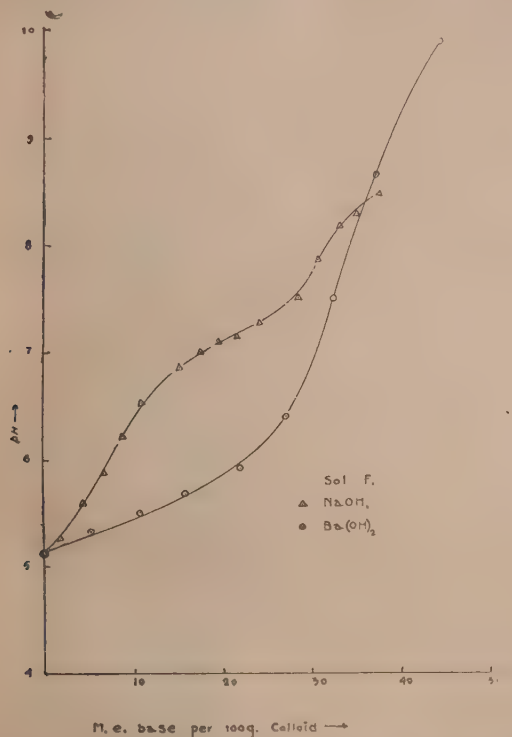


FIG. 1. Titration curves of hydrogen clay from Burdwan soil before removal of free oxides

The changes consequent on the removal of the free oxides may be summed as follows :

Chemical composition	SiO ₂ (per cent) $F_b > F$	Al ₂ O ₃ (per cent) $F_b > F$	Fe ₂ O ₃ (per cent) $F_b < F$
Base exchange capacity	$F_b < F$		
Form of titration curves	The curves of F_b show a weaker initial buffer action than those of F ; the $Ba(OH)_2$ curve of F_b shows a dibasic acid character not observed with F		

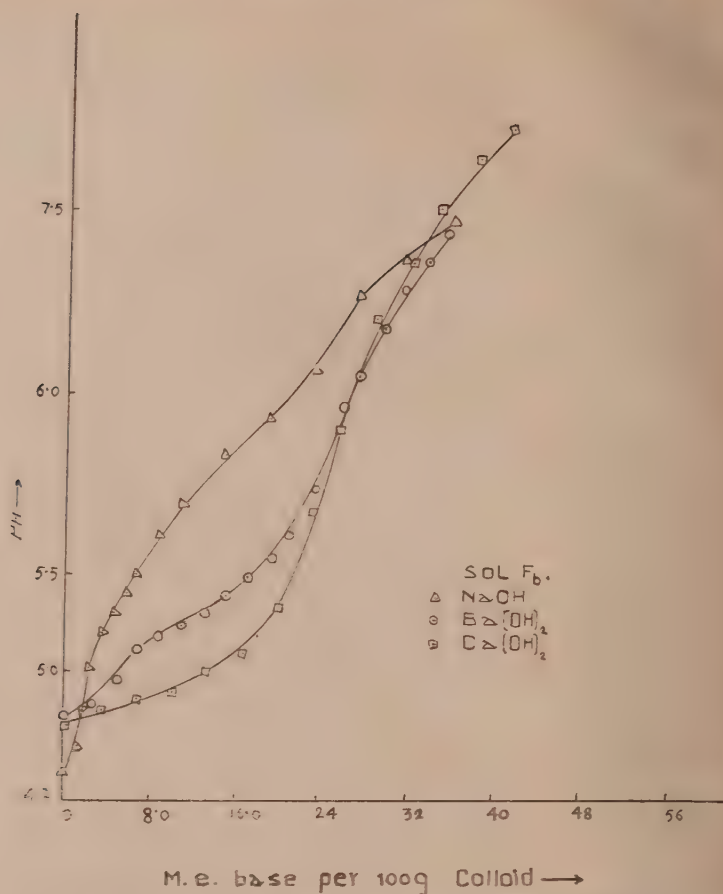


FIG. 2. Titration curves of hydrogen clay from Burdwan soil after removal of free oxides by Tamm's method

The alterations in chemical composition and the form of the titration curves point to a definite change in the hydrogen clay as a result of the treatment. The decrease in the b.e.c. suggests that this treatment brings about a decomposition of the exchange complex; an increase would have been observed if only free oxides having little or no base combining capacity had been removed.

B. Hydrogen clays prepared from the black cotton soil from Satara (Lab. No. 25) and the black soil from Raipur (Lab. No. 32)

The free oxides were removed by the method of Drosdoff and Truog. The base exchange capacities of I and K and their derivatives I_c and K_c calculated from the titration curves have been given in Table III and the results of fusion analysis in Table IV. All the four hydrogen clays give the same

type of titration curve with any given base. The titration curves of I_c only are given in Fig. 3.

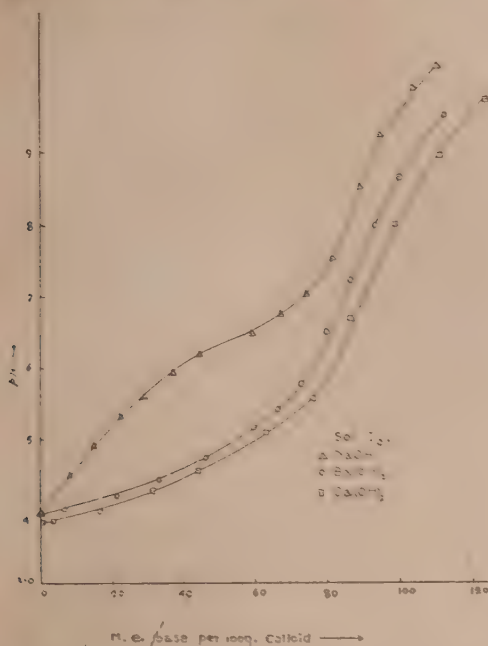


FIG. 3. Titration curves of hydrogen clay from the Satara soil after removal of free oxides by Drosdoff and Truog's method

TABLE III

Base exchange capacities of hydrogen clays from the Satara and Raipur soils before and after removal of free oxides by Drosdoff and Truog's method

Hydrogen clay	Base used for titration	pH at inflexion point in the titration curve	B.e.c. in m.e. base per 100 gm. oven-dried colloid	
			At inflexion point	At pH 7.0
I	NaOH	8.05	90.0	78.0
	Ba(OH) ₂	7.00	82.0	82.0
	Ca(OH) ₂	6.95	96.0	97.0
I_c	NaOH	8.10	86.0	74.0
	Ba(OH) ₂	7.60	91.0	85.0
	Ca(OH) ₂	6.50	86.0	91.0
K	NaOH	7.15	68.0	67.0
	Ba(OH) ₂	5.80	55.0	61.0
	Ca(OH) ₂	5.20	58.0	67.0
K_c	NaOH	8.10	67.5	60.0
	Ba(OH) ₂	5.75	56.0	62.0
	Ca(OH) ₂	5.78	63.0	68.0

TABLE IV
*Chemical compositions of hydrogen clays from the Satara and Raipur soils
 before and after removal of free oxides*

Hydrogen clay	SiO ₂ per cent	Al ₂ O ₃ per cent	Fe ₂ O ₃ per cent	SiO ₂ /R ₂ O ₃ (molar)	SiO ₂ /Al ₂ O ₃ (molar)
I . . .	55.03	24.20	20.81	2.50	3.87
Ic . . .	55.95	24.57	18.71	2.54	3.72
K . . .	55.93	28.03	14.80	2.54	3.40
Kc . . .	58.30	26.90	15.80	2.66	3.65

The changes consequent on the treatment are summed up in the chart given on the next page.

If the alterations in chemical composition merely indicated a removal of free oxides which are considered to have little base combining power, an increase in the b.e.c. would be expected. The b.e.c. of the Satara hydrogen clay actually shows a slight decrease* which probably indicates that a decomposition of the exchange complex is responsible for the variation in chemical composition. The b.e.c. of the other hydrogen clay remains practically unaltered and in order to reconcile this result with the small but definite variation in the chemical composition, it has to be assumed that the substances removed by the treatment were not 'inert'; they had, mass for mass, nearly the same b.e.c. as the original hydrogen clay.

C. Hydrogen clays prepared from the Bhata laterite soil (Lab. No. 33) and the red lateritic soil from Dacca (Lab. No. 22)

Laterite soils usually contain free sesquioxides. Mattson's method² was therefore used in the case of the above two hydrogen clays.

The b.e.c.'s of L and N and their derivatives L_a and N_a are given in Table V and the results of fusion analysis in Table VI. Figs. 4 and 5 show the titration curves of L and L_a. The titration curves of the other two hydrogen clays are similar to those of L and have been omitted.

*Except for the slight increase in the b.e.c. at pH 7.0 with Ba(OH)₂; this increase, however, is almost within the limits of experimental error (± 2.5 per cent)

	Chemical composition			B.e.c. with				Term of titration curves with bases	
				NaOH		Ba(OH) ₂			Ca(OH) ₂
	SiO ₂ per cent	Al ₂ O ₃ per cent	Fe ₂ O ₃ per cent	At inflexion point	At pH 7.0	At inflexion point	At pH 7.0		At inflexion point
Hydrogen clay from									
1. Black cotton soil from Satara	Increases	Increases	Decreases	Slightly decreases	Slightly increases	Increases but the pH at inflexion after removal of free oxides is definitely higher	Slightly increases	Decreases	The form of the curves remains practically unaltered except for variations in the pH's at the inflexion point
2. Black soil from Raipur	Increases	Decreases	Increases	No change though the pH at inflexion after removal of free oxides is higher	Decreases	No change	No change	Increases but pH at inflexion after removal of free oxides is higher	No change

The form of the curves remains practically unaltered for variations in the pH's at the inflexion point

Increases but pH at inflexion after removal of free oxides is higher

No change though the pH at inflexion after removal of free oxides is higher

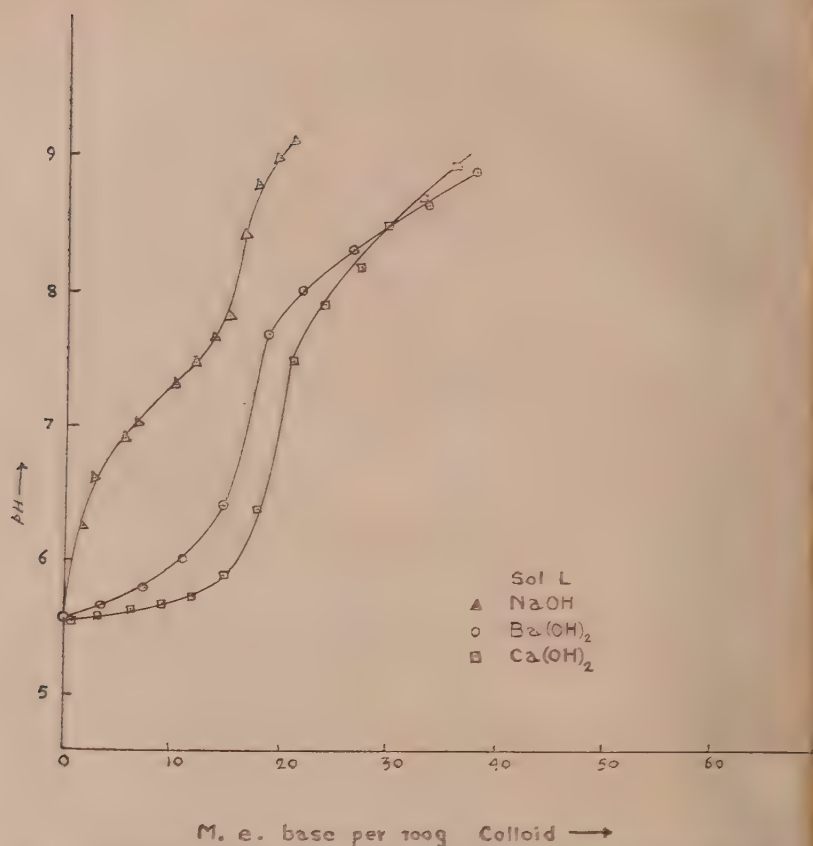


FIG. 4. Titration curves of hydrogen clay from Dacca soil before removal of free oxides

TABLE V

B.e.c. in m.e. base per 100 gm. of hydrogen clays from Dacca and Bhata before and after removal of free oxides by Mattson's method

Hydrogen clay	Base used					
	NaOH		Ba(OH) ₂		Ca(OH) ₂	
	At inflexion point	At pH 7.0	At inflexion point	At pH 7.0	At inflexion point	At pH 7.0
L . . .	16.25	6.3	17.5	17.0	19.0	19.0
L _a . . .	7* ; 6.0**	6.8	8.5	9.0	11.5	11.5
N . . .	18.	11.3	19.0	19.0	20.5	21.0
N _a . . .	11.	7.5	15.3	15.3	17.5	18.0

*Calculated from the first inflexion point

**Calculated from the second inflexion point

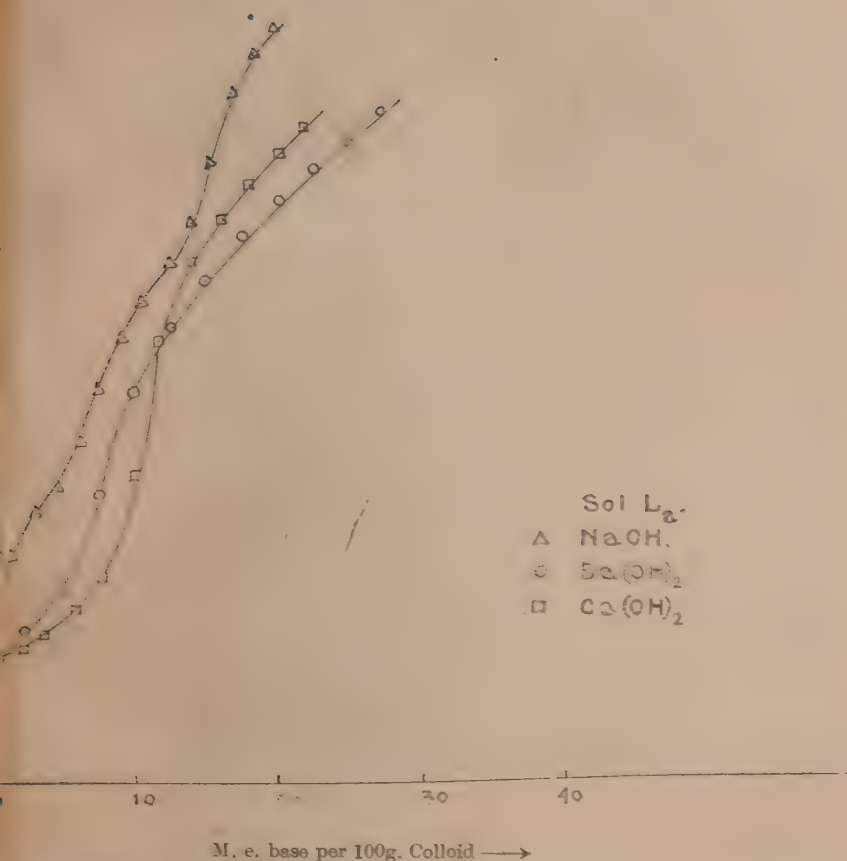


FIG. 5. Titration curves of hydrogen clay from Dacca soil after removal of free oxides by Mattson's method

TABLE VI

Chemical compositions of hydrogen clays from Dacca and Bhata soils before and after removal of free oxides

Hydrogen clay	SiO ₂ (per cent)	Al ₂ O ₃ (per cent)	Fe ₂ O ₃ (per cent)
• •	51.2	36.0	12.0
• •	50.0	38.8	10.1
• •	42.6	3.70	54.2
• •	42.0	4.50	53.4

The percentage of silica and ferric oxide decreases as a result of the treatment, while that of alumina increases. The b.e.c. of both L and N decreases. If only free sesquioxides having negligible b.e.c. were removed, an increase in the b.e.c. would have been observed. The treatment also brings about a material change in the form of the NaOH curve of L. While L behaves as a weak monobasic acid judging from this curve, its derivative L_a shows a dibasic acid character.

Attention may finally be drawn to one general feature observed with the hydrogen clays both before and after the treatments for the removal of their free oxides. At any given pH, the slopes of their titration curves with different bases are usually arranged in the order $\text{NaOH} > \text{Ba(OH)}_2 > \text{Ca(OH)}_2$ which shows that these three bases react with the hydrogen clays in the reverse order. This difference in the relative effects of the three bases illustrates what has been designated by us as an irregular or specific cation effect. This has been fully discussed elsewhere [Mitra, 1936, 1940; Mukherjee, Mitra and Mukherjee, 1937].

SUMMARY

A study has been made of the effect of treatments aiming at the removal of free silica and sesquioxides contained in hydrogen clays on their chemical composition, nature of titration curves with bases and base exchange capacity (b.e.c.). Hydrogen clays prepared from the entire clay fraction of five Indian soils have been used and the methods of Tamm, Drosdoff and Truog, and Mattson were employed for the removal of the free oxides. Tamm's method gives rise to a decrease in the b.e.c. of a hydrogen clay from a Burdwan (Bengal) soil. The chemical composition and the form of the titration curves are materially altered indicating, on the whole, a decomposition of the exchange complex as a result of the treatment. Practically no change occurs in the b.e.c. and the nature of the titration curves of hydrogen clays from two black cotton soils using the method of Drosdoff and Truog. Slight changes in the chemical composition are, however, observed. Mattson's method gives rise to a marked decrease in the b.e.c. of hydrogen clays from a red lateritic soil from Dacca (Bengal) and a red laterite soil from the Central Provinces. The form of the titration curve with caustic soda of the hydrogen clay from the Dacca soil is also altered.

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STUDIES IN KUMAUN HILL SOILS

III. SOIL TYPES AT DOONAGIRI

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THE soil survey work reported by us for Chaubattia in the first two parts of this series of publication has also been extended to other parts of Kumaun, and in the present paper a consolidated account of the soil types found at Doonagiri is discussed. The technique of our survey and the methods employed have already been described in detail in the previous papers, and in the present instance only a bare outline of these will be given.

Doonagiri lies north of Chaubattia at a distance of 16 miles as the crow flies.

The parent material of the soils is phyllite; granite gneiss has been found at only a few places. Although a large number of soils studied by us are clayey, due to high content of organic matter, the soils, however, are not waterlogged. The general forest flora of the locality is very much similar to that of Chaubattia. Average rainfall is over 60 inches a year. Situated further interior into the hills, and being a little higher, the climate of Doonagiri is slightly cooler and comparatively more humid than that of Chaubattia. The estate was deforested in about 1867 and planted to tea. After about 70 years due to transport and other difficulties the plantation was abandoned, and no attempt has since been made to rehabilitate the area.

LITERATURE

Information on hill soils is very scanty and the literature suffers from lack of data. The present position of the work on hill soils was summarized in the two foregoing papers of this series [Mukerji and Das, 1940; 1941], and an attempt has, therefore, been made here to review the literature.

The importance of the clay fraction in soil characterization was shown by Benson [1930] for certain profiles of north Wales. Various authors have since utilized this method for the classification of soil profiles studied by them. Recently Mukerji and Das [1940; 1941] in their studies on hill soils have shown the usefulness of $\text{SiO}_2/\text{R}_2\text{O}_3$ ratio in determining the precise characteristics of soil types in the Himalayan foot-hills in the United Provinces.

ANALYTICAL METHODS

Clay

International pipette method was used for the determination of content of soils. After preliminary treatment with hydrogen peroxide hydrochloric acid, dispersion was brought about by ammonia.

Organic carbon

Walkley and Black's method [1934] was used for this determination.

pH

pH values were determined by means of quinhydrone electrode.

Sesquioxides

For the determination of sesquioxides, HCl digestion was done according to the method of Agricultural Education Association.

Base-saturation

Barium acetate and ammonium chloride method of Parker [1929] utilized for the determination of exchangeable acidity and base-exchange capacity in the same sample.

Clay composition

Robinson's method [Wright, 1939] was followed, and sesquioxides silica were determined by the usual method for estimating silicates.

DATA AND DISCUSSIONS

A large number of soil profiles were examined at Doonagiri, and in succeeding pages a critical account of six profiles typical of the area will be given. It has been shown in the previous two papers of this series that silicic acid and sesquioxide ratios together with the figures for percentage base-saturation give more or less an accurate idea of the local soil types. These data will therefore, be examined for all the profiles studied along with such other relevant data as necessary.

Pit No. 2 : Doodhatoli

Horizon	Depth	Description
I . . .	0-1 ft. 3 in. . .	Grey ; granular ; loamy with a slightly bluish tinge. Dark grey when wet
II . . .	1 ft. 3 in.-2 ft. 3 in.	Same as above ; particles more granular appear to contain more organic matter than above
III . . .	2 ft. 3 in.-4 ft. 6 in.	Organic ; clayey loam ; dark grey. Colour same as of the second horizon

TABLE I
Summarized analytical data of Doonagiri soils
(Pit No. 2)

Horizon	Per cent (air-dry basis)					C/N	pH
	Clay	Organic carbon	Total nitrogen	Fe ₂ O ₃	Al ₂ O ₃		
I	23.35	3.9	0.23	5.59	8.97	16.96	6.2
II	21.75	7.6	0.25	8.32	8.10	30.40	5.8
III	30.50	5.8	0.20	8.48	7.67	29.00	5.5

The general character of the profile is organic, and Wiesenboden characteristics are revealed in C/N ratios. Clay content of different horizons, and sesquioxide distributions are rather erratic. The composition of clay fraction and base-saturation percentages of different horizons are given in Table

TABLE II
Clay analysis of Doonagiri soils
(Pit No. 2)

Horizon	Per cent (air-dry basis)			SiO ₂ R ₂ O ₃	Exchangeable H (m. e. per cent on air-dry basis)	Per cent base- saturation
	SiO ₂	Al ₂ O ₃	Fe ₂ O ₃			
I	44.48	19.88	15.97	2.51	4.21	75.56
II	43.90	20.93	16.77	2.35	7.25	54.69
III	43.20	26.23	16.37	2.00	8.70	54.69

These figures, particularly eluviation of Al₂O₃ and constancy of Fe₂O₃ in the profile, indicate that the third horizon is not of the same age as the first two, and can be classified as a brown forest soil having podsollic tendencies.

Pit No. 3 : Doodhatoli

Horizon	Depth	Description
I	0-11 in.	Grey; granular; loamy; dark grey when wet
II	11 in.-1 ft. 10 in.	Dark grey; clayey; loamy; very dark grey when wet
III	1 ft. 10 in.-4 ft.	Dark grey; granular; loamy; very dark grey when wet

TABLE III
Summarized analytical data of Doonagiri soil
(Pit No. 3)

Horizon	Per cent (air-dry basis)					C/N	pH
	Clay	Organic carbon	Total nitrogen	Fe ₂ O ₃	Al ₂ O ₃		
I	24.45	5.5	0.30	4.95	13.07	18.33	6.0
II	29.25	6.4	0.23	8.24	10.49	27.83	5.7
III	22.50	4.8	0.14	8.80	13.36	34.29	5.9

The predominant character of the entire profile is its organic matter content, and C/N ratio increases with depth. In this respect the pedologic characters of the two profiles examined at Doodhatoli are alike. Clay content and pH values are rather erratic. Clay analysis figures of this profile and base status are given in Table IV.

TABLE IV
Clay analysis and exchangeable bases of Doonagiri soils
(Pit No. 3)

Horizon	Per cent (air-dry basis)			SiO ₂ R ₂ O ₃	Exchangeable H (m.e. per cent on air-dry basis)	Per cent base-saturation
	SiO ₃	Al ₂ O ₃	Fe ₂ O ₃			
I	45.74	18.23	15.57	2.75	7.98	65.33
II	43.86	25.43	15.97	2.10	7.25	60.39
III	42.08	27.63	17.97	1.82	5.80	67.78

It follows from figures given above that the second and third horizon show some of the typical characters of the brown forest soils having Wiese boden podsolic tendencies. The first horizon, however, does not appear to be of the same age. A few more profiles were examined in this area and visual characters they were found to be similar to pit Nos. 2 and 3.

Pit No. 5 : Dadoi

Horizon	Depth	Description
I	0-10 in.	Grey ; granular ; loamy ; dark grey when wet
II	10 in.-1 ft. 5 in.	Grey with a slightly brownish tinge ; loamy ; contains more clay than the top ; more grey when wet
III	1 ft. 5 in.-3 ft.	Brownish ; granular ; clayey loam ; more brown when wet

TABLE V
Summarized analytical data of Doonagiri soils
(Pit No. 5)

Per cent (air-dry basis)					C/N	pH
Clay	Organic carbon	Total nitrogen	Fe ₂ O ₃	Al ₂ O ₃		
25.40	2.81	0.18	5.35	13.63	15.61	6.0
32.85	1.9	0.15	8.32	8.39	12.62	5.6
36.50	1.4	0.08	8.48	10.28	17.50	5.4

It is clear from the above description and analytical data that these profiles are heavy brown forest soils, undisturbed by terracing operations, and very much affected by surface erosion. The composition of the clay fractions, as given in Table VI, indicates the real nature of the profile.

TABLE VI
Clay analysis and exchangeable bases of Doonagiri soils
(Pit No. 5)

Horizon	Per cent (air-dry basis)			SiO ₂ R ₂ O ₃	Exchangeable H (m.e. per cent air-dry basis)	Per cent base-saturation
	SiO ₂	Al ₂ O ₃	Fe ₂ O ₃			
I	43.50	29.63	15.57	1.86	2.18	86.41
II	46.26	26.93	14.77	2.16	4.35	74.71
III	47.60	25.03	13.57	2.40	9.43	36.32

Although there is a slight indication of eluviation of silica, exchangeability and percentage base-saturation figures clearly bring out the brown forest soil characteristics of the profile. The silicious material of the second and third horizons has sometimes been ascribed to colloidal SiO₂ of the parent material. It is suggested that the excess of silica in the second and third horizons of most of our soils is due to capillary ascendance of silica soil from lower layers.

No. 7 : Khalkhet

Horizon	Depth	Description
I	0-6 in.	Grey ; granular ; loamy sand with a slightly brownish tinge with some clay ; dark grey when wet
II	6 in.-1 ft. 6 in.	Same as above, but contains more clay
III	1 ft. 6 in.-2 ft. 6 in. and below.	Brownish grey ; more granular, heavy loam

TABLE VII
Summarized analytical data on Doonagiri soils
(Pit No. 7)

Horizon	Per cent (air-dry basis)					C/N	pH
	Clay	Organic carbon	Total nitrogen	Fe ₂ O ₃	Al ₂ O ₃		
I	13.25	6.4	0.22	7.19	7.23	29.09	6.5
II	37.50	4.2	0.15	8.88	8.29	28.00	6.0
III	35.25	4.6	0.14	10.08	9.19	32.86	5.8

The first 6 in. of the profile have been impoverished of its finer material presumably by erosion. In all other essential characters, these profiles resemble brown forest soils. The essential brown forest characters of the profile brought out more clearly from the clay analysis figures given in Table VII.

TABLE VIII
Clay analysis and exchangeable bases of Doonagiri soils
(Pit No. 7)

Horizon	Per cent (air-dry basis)			SiO ₂ R ₂ O ₃	Exchangeable H (m.e. per cent on air-dry basis)	Per cent base saturation
	SiO ₂	Al ₂ O ₃	Fe ₂ O ₃			
I	47.22	27.43	14.37	2.20	5.08	70.0
II	45.16	28.03	15.17	2.03	4.35	72.4
III	44.90	28.43	14.77	2.01	7.25	59.5

Whereas the character and composition of exchangeable bases resemble those of brown forest soils, a slight podsollic tendency is indicated by the low in SiO₂/R₂O₃ ratios.

Pit No. 6 : Khalkhet (Lower level of southern highlands)

Horizon	Depth	Description
I	0-6 in.	Greyish brown ; loam containing undecomposed organic matter ; more brown when wet
II	6 in.-1 ft. 5 in.	Yellowish brown ; heavy loam ; more brown when wet
III	1 ft. 5 in.-2 ft. 6 in.	Reddish brown ; heavy loam ; deep red when wet

TABLE IX

Summarized analytical data on Doonagiri soils

(Pit No. 7)

Horizon	Per cent (air-dry basis)					C/N	pH
	Clay	Organic carbon	Total nitrogen	Fe ₂ O ₃	Al ₂ O ₃		
I	28.95	2.7	0.13	4.67	17.23	20.85	6.4
II	32.75	1.0	0.07	12.48	10.24	14.29	5.8
III	32.75	0.8	0.05	14.96	14.46	16.00	5.7

There is quite an appreciable eluviation of clay; pH values and organic carbon percentages resemble those of brown forest soils. We find, however, considerable translocation of ferric oxide, which, as is well known, is a podsollic character.

The results of the clay analysis and data on exchangeable bases are presented in Table X.

TABLE X

Clay analysis and exchangeable bases of Doonagiri soils

(Pit No. 6)

Horizon	Per cent (air-dry basis)			SiO ₂ R ₂ O ₃	Exchangeable H (m.e. per cent on air-dry basis)	Per cent base-saturation
	SiO ₂	Al ₂ O ₃	Fe ₂ O ₃			
I	44.04	26.83	16.77	1.99	8.70	55.84
II	44.60	24.03	15.97	2.21	1.45	87.50
III	40.62	21.34	20.76	1.99	1.45	89.10

The data given above leave no doubt as to the genetic nature of these profiles. There is a very high eluviation of Fe₂O₃, specially from the first to third horizon. The base-exchange figures also indicate podsollic tendency of the profile under reference. In view of the constancy of composition of SiO₂/R₂O₃ ratios this profile may be classified as brown forest soil with podsollic tendency.

The general characters of all the five profiles discussed above show the existence of one genetic type of soil formation in this area. A casual observation, however, of the hill slopes in this area as well as over the whole of Kumaun

shows the presence of a large variety of soils with colours varying from red to yellow. These soils are often shallow and occupy quite an appreciable area of the Kumaun hills, and are utilized for the growing of grain or fruit trees. A number of profiles were examined in this area and such profile is described under and summarized analytical data are presented in Table XI.

Pit No. 1 : Kiyari (Western ridge)

Horizon	Depth	Description
I	0-1 ft. 6 in.	Yellowish brown; loamy sand; more brown when wet and sticky. Some decomposed platy phyllite rock present

TABLE XI

Summarized analytical data of Doonagiri soils

(Pit No. 1)

Per cent (air-dry basis)					C/N	pH
Clay	Organic carbon	Total nitrogen	Fe ₂ O ₃	Al ₂ O ₃		
16.45	0.86	0.03	8.70	20.50	28.66	5.0

Absence of organic matter and therefore the seat of eluvial activity is clearly indicated by both visual observations and by low contents of organic carbon and total nitrogen. The figure for the sesquioxides is comparatively higher and pH value much lower than what is usual for eluvial horizon in this locality.

Data on clay composition and exchangeable bases are given in Table XII.

TABLE XII

Clay composition and exchangeable bases of Doonagiri soils

(Pit No. 1)

Per cent (air-dry basis)				Exchangeable H (m. e. per cent on air-dry basis)	Per cent base-saturation
SiO ₂	Al ₂ O ₃	Fe ₂ O ₃	$\frac{\text{SiO}_2}{\text{R}_2\text{O}_3}$		
45.02	20.47	20.73	2.27	4.99	62.48

It is clear from the data presented in Tables XI and XII that the profile has some essential characters of lower horizons of pure brown forest soils discussed earlier in this section. The predisposing cause leading to the development of these soils is intense surface erosion in the past, but often such conditions are also brought about by faulty terracing practices. The soils because of their acidic nature and peculiar dynamics should be classified as a variety of brown forest soils resembling truncated forest soils of Sigmond's [1938] main type 4 of soil order 10. Other profiles studied in this area have characteristics similar to those described in these pages and can be classified into two major genetic groups, viz. brown forest soils and brown forest soils with podsollic tendency.

DISCUSSION

In the first two parts of this series of papers the possibility of classifying hill soils according to their developmental characteristics has been clearly indicated. In the present instance the same method of soil classification has been followed. It has been possible by this method in the present case not only to understand the characteristics of hill soils as a class but also the genesis of the individual profiles. It is interesting to note that silica/sesquioxide ratios of the clay fractions and per cent base-saturation figures taken together supply all the important data necessary for the classification and characterization of soils in the hills. It is not implied by this that the organic matter content, pH and mechanical composition do not offer any valuable assistance in the studies of hill soils, but that such determinations have limited importance in consequence of the fact that they only confirm the findings arrived at from study of the clay composition and exchangeable bases.

Doonagiri soils belong to the usual brown forest soil type and on the average these soils are very much richer in organic matter. Moreover, the soils of this locality contain a high proportion of plant nutrients. The largest number of successful fruit orchards of these hills are situated on soils formed on phyllite rock like that of Doonagiri, a fact that cannot be ascribed to chance alone.

The prevalent colours of most of these soils at Doonagiri are black and reddish brown. Soils having the latter colour, as has been indicated above, belong to truncated brown forest soil type. Soils having black colour are an important study by themselves. It is not, as we find, that the higher content of organic carbon always accounts for the black colour of the soil, but a high C/N ratio seems to have some direct bearing on the black colouration. Such seems to have been the case with pit No. 2. Soil containing 7.6 per cent of organic carbon appears to be as organic as another soil which contains only 3.8 per cent organic carbon. Pit No. 3 also shows similar characteristics. Profile No. X 15 Y 15 [Mukerji and Das, 1941] is an analogous case.

The mean $\text{SiO}_2/\text{R}_2\text{O}_3$ ratio in the clay fractions of these profiles is 1.15 ± 0.062 . This ratio is well within limits to be consistent with the hypothesis that the primary weathering product of this locality is a mixture of hydrated silicates having the general formula $\text{R}_2\text{O}_3 \cdot 2\text{SiO}_2 \cdot x\text{H}_2\text{O}$. Deviations from absolute value of 2 may have been brought about by eluviation and translocation. The usefulness of the silica/sesquioxide and silica/

alumina ratios for determining with certainty the genetic characterisation of hill soils will be discussed more fully in the subsequent parts of this series of publications dealing with the developmental characters of soils derived from other rock materials.

SUMMARY

The nature of some soils formed at Doonagiri has been discussed.

Clay composition and per cent base-saturation taken together have been found very useful in characterizing these soils.

Silica/sesquioxide ratio has been found to be 2.15 ± 0.24 and, therefore, the primary weathering product has been suggested to be a clay having the general composition $R_2O_3 \cdot 2SiO_2 \cdot xH_2O$.

The soils of the locality contain large proportions of organic matter, and surface soils are on the whole highly base-saturated.

From the general study of the data it is evident that these soils belong to the brown forest soil group.

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STUDIES IN INDIAN RED SOILS

VI. DETERMINATION OF MINERALOGICAL COMPOSITION

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(With one text-figure)

The usual method of characterizing soil types in the laboratory is to find out the physical and chemical composition of the soil samples, as also of the clay fractions. These determinations, however, require long time to be carried out. Comparatively little attention has been paid, so far to the determination of the mineral compositions of soils, at any rate in India. The presence of great variety of minerals in soils was noted by McCaughey and Fry [1913], who reported the results of mineralogical studies on the chief soil groups of the United States. They identified optically 34 different minerals in a great variety of soils and concluded that the mineralogical composition of soils varies with the physiographic regions in which they occur. Among other workers who have discussed the constitution of soils from mineralogical standpoint are Delage and Lagatu [1904 ; 1905], Cayeau [1905], Hendrick and Newlands [1923; 1925 ; 1928], Hart [1935], Marshall [1935 ; 1936], Pearson and Truog [1937] and Bonnett [1939]. The determination of soil minerals is useful to the study of soils in two different ways :—

(1) In the first place, the content of minerals may indicate the nature of the parent material of the soil.

(2) In the second place, the determination of soil minerals may indicate the nature of inorganic soil colloids and of plant foods present in the soil. Moreover, permanence of fertility in a soil varies with its contents of minerals which are still liable to decomposition. A soil having a high content of such minerals is, other things being equal, better than a soil with no mineral reserves [Vageler, 1933].

In connection with his study on the nature of colloidal minerals of clay, Nagelschmidt [1939] has discussed all the important existing methods for determining the nature of clay minerals, viz. X-ray, optical, dehydration and chemical methods with special regard to their limitations. On the other hand, the importance of determining minerals of microscopic dimensions by the petrological methods has been pointed out by various workers (for a survey of literature see Harrison [1933]). In view of the scanty data on the mineralogical composition of Indian soils, it was felt desirable to determine the mineralogical composition of fine sand fractions of some typical profile

samples of red soils collected from different parts of India. The composition with respect to the rock-forming minerals of the soil is well represented by the fine sand fraction, except for minerals with flaky structure, such as mica, which tend to accumulate in the finer fractions. Volk [1933], in studying the formation of muscovite in soils, reports quantitative separation of mineral groups by means of liquids of different specific gravities. In recent papers, Truog and coworkers [1937-38] have sub-divided mineralogical constituents of soil by means of specific gravity separation with heavy liquid, such as tetra-bromo-ethane and nitro-benzene mixtures. Hendrick and Newlands [1923] have obtained good results on the separation of the heavy and light fractions of fine sand by bromoform (sp. gr. 2.9). Also by using a mixture of bromoform and benzene (sp. gr. 2.65), they separated quartz particles from the felspar grains, the sp. gr. of the quartz particles being higher than 2.65 (middle fraction), whilst that of the felspar particles being less (light fraction).

EXPERIMENTAL

The method of separation of the heavy, middle and light fractions of the fine sand, for the mineralogical and microscopical examination was essentially that described by Hendrick and Newlands [1923]. The work in the present paper can be divided into two sections. In the initial stages of the work (§I) the fine sand fractions with which the microscopical determinations were carried out, were not estimated quantitatively for the percentages of heavy, middle and light fractions. At this stage of the work, the heavy and light fractions were separated by shaking with bromoform (sp. gr. 2.9) only, but the separated fractions were not weighed. Also the nature of minerals on the microscopic slides were only qualitatively determined, but they were not counted for determining the percentages of different minerals in the slides.

In the second stage of the work (§II) it was felt desirable to obtain the mineralogical data of the soil samples on a quantitative basis. The fine sand fractions were accordingly separated into three fractions (heavy, middle and light) and their composition separately determined with the help of petrological microscope. The percentages of different minerals present in the slide were also carefully counted with the help of a graduated cross-eye-piece micrometer scale.

Separation of fine sand into heavy and light fractions

A special separating funnel illustrated in Fig. 1 was used to facilitate the work and minimize the chances of admixture of the separated minerals. The funnel is provided with a stopper closed at one end. The separated heavy mineral, therefore, collects in the opening so that by turning out the stopper the separation is complete. For actual experiment approximately 1.5-2 gm. of fine sand, after ignition, were treated with approximately 20 c.c. of *N/10* oxalic acid and the mixture kept in contact for about 16 hours. By this treatment the grain were cleared of ferruginous matter. The supernatant liquid was then thrown out and the residue washed 10 times with distilled water by decantation in a beaker. The substance was then dried and an

curately weighed portion of it was treated with sufficient quantity of bromoform in the special separating funnel described before. It was necessary for the bromoform and the substance to remain in contact with each other for a period of not less than three to four hours. The heavy fraction was then isolated by turning out the stopper and was transferred to a dry filter paper, washed with benzene and dried (heavy fraction). The light fraction which was floating on the surface of the bromoform was then filtered through dry filter paper, washed with benzene and dried. This dried substance was subsequently treated in the same separating funnel with a mixture of bromoform and benzene of sp. gr. 2.62. The fraction which settled at the bottom of the separating funnel and that which floated on the surface of the liquid were collected separately on dried filter papers, washed with benzene and dried (middle and light fractions).

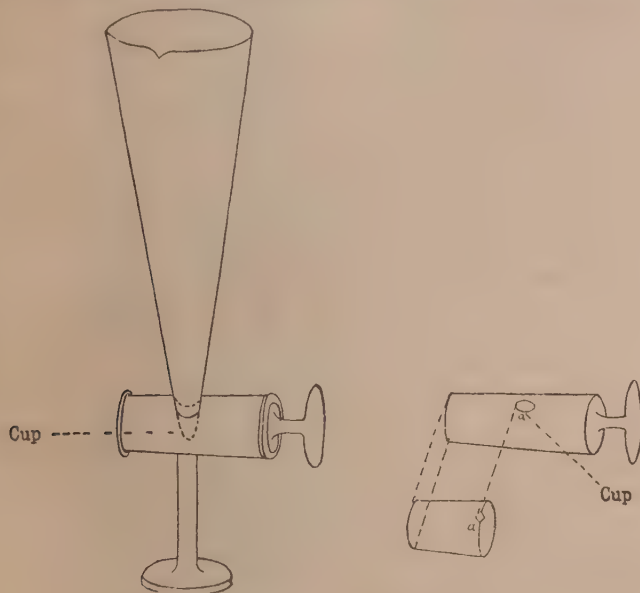


FIG. 1. Spaeth's separating apparatus

The sample and liquid are placed in the funnel and stirred. The heavy residue collects in a cup in the tap: the turning of the tap isolates the residue until the light minerals have been removed)

Preparation of microscopic slides

The separated fine sand fractions were mounted on microscopic slides with Canada balsam by following essentially the procedure described by Milner [1932].

§I. RESULTS AND DISCUSSION

The results have been summarized in Table I and, as far as possible, the minerals have been written in descending order of abundance. It will be found that iron ore minerals (e.g. limonite, magnetite, and haematite), as

also quartz and felspar grains, are uniformly present in high concentrations in all the profiles.

TABLE I

Soil minerals at different localities and the geological formations of those localities

Locality	Minerals	Parent materials*
Dacca Farm, Bengal	Iron ore minerals, quartz and felspar grains, epidote, hornblende, zircon and chloritic minerals	Old alluvium + ?
Suri, Birbhum, Bengal	Iron ore minerals, quartz and felspar grains, epidote, zircon, garnet and chloritic minerals	Recent deposit + ?
Bidar, Hyderabad	Iron ore minerals, quartz and felspar grains, garnet, chloritic minerals, epidote and biotites	Basalt + ?
Himayethsagar, Hyderabad, Deccan	Iron ore minerals, quartz and felspar grains, hornblende, epidote, augite and chloritic minerals	Granite + ?
Telankheri, Nagpur, C. P.	Iron ore minerals, quartz and felspar grains, epidote, zircon and chlorites and occasionally augites and hornblende. Also a few biotites	Basalt + ?
Chandkhuri Farm, Raipur, C. P.	Iron ore minerals, quartz and felspar grains, garnet, zircon, chloritic minerals	Limestone + ?
Alisagar, Hyderabad	Iron ore minerals, quartz and felspar grains, epidote, garnet, augites and a few chloritic minerals	Quartzite + ?
Kakat, Cannanore, Malabar	Iron ore minerals, quartz and felspar grains, epidote, zircon, staurolite and chloritic minerals	Granite + ?
Puzathi, Cannanore, Malabar	Iron ore minerals, quartz and felspar grains, epidote, augite, zircon and chlorites, occasionally tourmalines	Granite + ?
Nilgiri Hills, Madras province	Iron ore minerals, quartz and felspar grains, epidote, augite, garnet, chlorite, zircon and biotite	Charnolite + ?
Gorantla Hills, Guntur, Madras	Iron ore minerals, quartz and felspar grains, epidote, zircon, garnet, hornblende and a few staurolites	Granite + ?
Stambhalaguruva, Guntur, Madras	Iron ore minerals, quartz and felspar grains, epidote, zircon, garnet, staurolites and chlorite, occasionally a few topaz	Granite + ?
Khodappanam Kunnu, Trivandrum, Travancore	Iron ore minerals, quartz and felspar grains, epidote, zircon, garnet, chloritic minerals and a few muscovites	Gneiss + ?
Government Fruit Farm, Cape Comorin	Iron ore minerals, quartz and felspar grains, epidote, hornblende, zircon, tourmaline and chloritic minerals	Gneiss + ?

* It is likely that the exposed rocky pieces, which were collected from the places where the profiles were taken, constitute a part of the parent material. Hence the sign ' ? ' has been added.

Many of the potash feldspars of smaller sizes were turbid with decomposition products (kaolinization). In almost all the soils the quartz particles characterized by rough angular appearance. Rounded grains are also sometimes met with. Table I shows that the common ferro-magnesian rock-forming minerals are represented by one or more species in all the soils examined, one or other being present in preponderating amount. Hornblende is present in some cases and that too in small amounts. Chlorite, an alteration product from the ferro-magnesian rocks was found in most of the soils studied. Epidote was occasionally found (e.g. Nilgiri hills). These minerals are of resistant nature and they are concentrated in material which has been subjected to prolonged weathering and has accumulated from a variety of sources. Epidote and garnet were frequently found in granular form, whilst tourmaline, especially zircon, though sometimes fragmentary, showed crystalline structure.

Iron ore minerals like haematite, limonite and magnetite were present in all the soils examined. With regard to the percentage of the ferro-silicates and other minerals in various soil samples, the soils of basic igneous origin were found to be the richest. In almost all cases the proportion of resistant minerals like epidote, garnet, zircon and tourmaline is higher compared with common rock-forming minerals like biotite and hornblende. The latter minerals are the potential sources of plant food supply and these are usually present in the soils in small amounts, which explains the comparatively little productivity of these soil types. In a recent paper, Bonnett [1939] has examined the mineralogical constituents of the silt fractions of samples from a lateritic soil profile of Puerto Rico and has shown that the constituent minerals are hydrated iron oxides, gibbsite, muscovite, secondary quartz and serpentine. The accessory minerals, on the other hand, are : rutile, ilmenite, leucoxene, zircon, magnetite, chlorite, sericite, calcite, glass, and a kaolin-like mineral and traces of talc and epidote. The lateritic soils of India studied differ from those of Puerto Rico in that they are mostly found to occur on quartzites and granitic rocks except in the case of central and western and north-eastern parts of peninsular India, where the parent material is basalt. The parent material of soil type of Puerto Rico is Andesitic. The quartz particles found in such soil types seem to be mainly secondary quartz.

The presence in almost all the soil types of a large proportion of iron minerals and highly resistant unweathered minerals such as epidote, zircon, tourmaline, etc. suggests that the soil has been formed of prolonged chemical weathering which took place prior to present soil-forming processes. The alternative suggestion is that these soils have been subjected to extensive erosion and minerals of comparatively low specific gravity have been washed down behind minerals of comparatively high specific gravity like epidote, zircon, tourmaline, etc.

§ II. MECHANICAL ANALYSES OF SOIL SAMPLES

The mechanical analyses were carried out by following essentially the procedure developed by Robinson [1933]. The data on the percentages of coarse sand, fine sand, silt and clay are shown in Table II.

TABLE II
Mechanical composition of soil samples
 (Air-dry basis)

Locality	Sample No.	Depth	Coarse sand (per cent)	Fine sand (per cent)	Silt (per cent)	Clay (per cent)
Hathwara, Bihar	Manbhum,	81p 0—1 ft. 6 in. . .	38·0	21·0	9·0	30·0
		82p 1 ft. 6 in.—2 ft. 3 in. .	41·0	17·5	10·8	28·0
		83p 2 ft. 3 in.—3 ft. 6 in. .	34·9	21·6	10·8	31·0
		84p 3 ft. 6 in.—4 ft. 11 in. .	34·9	11·8	15·9	33·0
		85p Below 4 ft. 11 in. .	40·0	20·0	11·2	28·0
Putida, Singhbhum, Bihar		86p Below 30 ft. .	61·6	17·1	5·5	14·0
		87p 0—1 ft. . .	22·7	21·5	17·2	37·0
		88p 1 ft.—2 ft. 9 in. .	30·9	12·0	13·2	37·0
Ratu, Ranchi, Bihar		89p 2 ft. 9 in.—4 ft. .	48·8	15·5	10·4	19·0
		90p 0—1 ft. . .	23·8	19·6	14·9	37·0
		91p 1 ft.—2 ft. . .	17·9	14·5	15·4	48·0
		92p 2 ft.—3 ft. . .	26·0	13·0	13·2	44·0
		93p 3 ft.—4 ft. . .	31·3	11·3	9·1	45·0
Baralota, Bihar	Daltonganj,	94p 0—1 ft. 11 in. .	21·4	32·6	16·0	28·0
		95p 1 ft. 11 in.—2 ft. 9 in. .	15·7	21·1	13·8	44·0
		96p 2 ft. 9 in.—4 ft. .	16·9	16·0	12·6	49·0
		97p 4 ft.—5 ft. . .	48·8	17·1	19·6	12·0
Tangi, Cuttuck, Orissa		98p 0—1 ft. . .	35·0	33·8	14·8	14·0
		99p 1 ft.—2 ft. . .	32·1	27·9	17·0	20·0
		100p 2 ft.—4 ft. . .	49·1	20·1	9·1	17·0
Dhanmandal, Cuttuck, Orissa		101p 0—5 in. . .	28·7	23·8	14·6	31·0
		102p 5 in.—4 ft. . .	16·0	9·0	7·5	61·5
Kapileswar, Orissa	Bhubaneswar,	103p 0—2 ft. 11 in. .	40·1	21·0	11·6	26·4
		104p 2 ft. 11 in.—4 ft. .	49·0	19·4	3·1	27·6
		105p Below 30 ft. . .	41·3	15·7	12·6	28·0
Jhinkartangi, Khurda Town, Orissa		106p 0—1 ft. . .	34·3	10·2	12·2	39·4
		107p 1 ft.—2 ft. . .	22·1	11·0	13·4	48·9
		108p 2 ft.—8 ft. 6 in. .	44·7	11·7	11·7	28·3
		109p 8 ft. 6 in.—10 ft. .	42·5	24·9	6·7	22·4
		110p 30 ft.—50 ft. . .	29·9	20·5	19·0	29·3
Lalgah, Midnapur, Bengal		112p 0—4 in. . .	28·3	18·8	10·4	39·0
		113p 4 in.—3 ft. 4 in. .	40·0	18·5	2·8	35·2
		114p 3 ft. 4 in.—4 ft. .	46·6	18·2	7·6	26·7
		115p 7 ft.—8 ft. . .	13·4	41·5	6·6	34·2
Mailda, Midnapur, Bengal		116p 0—8 in. . .	39·3	44·7	4·4	9·5
		117p Bed soil of Cossye R. .	5·0	19·3	24·3	44·0
		118p 40 ft. below . .	39·5	13·4	13·2	32·8

TABLE II—*contd.*

Locality	Sample No.	Depth	Coarse sand (per cent)	Fine sand (per cent)	Silt (per cent)	Clay (per cent)
Jhalang, Khasi Hills, Assam	119p	0—6 in.	17·9	12·0	23·7	37·0
	120p	6 in.—1 ft. 3 in. .	20·5	11·3	23·3	35·6
	121p	1 ft. 3 in.—2 ft. 1 in. .	18·6	11·7	23·3	38·6
	122p	2 ft. 1 in.—4 ft. .	25·0	9·5	23·4	35·4
Chandmari, Tura, Jhalang Hills, Assam	144p	0—3 in.	43·5	18·4	14·7	20·2
	145p	3 in.—1 ft. 8 in. .	54·6	18·6	10·5	15·0
	146p	1 ft. 8 in.—2 ft. 8 in.	65·2	16·4	8·4	9·1
	147p	2 ft. 8 in.—4 ft. .	63·7	12·9	13·8	8·1

The mechanical analyses data show generally a more sandy texture of the surface soil than for the subsoil. This fact has been usually ascribed to a sandy wash of external origin, or to mechanical eluviation within the soil. Robinson [1936] has suggested a third possibility in that it might be due to normal erosion, as distinct from catastrophic erosion, involving lateral movement of the finer fractions. Such removal, according to him, might take place along the surface of the soil itself or along the surface of a parent material, and, in either case, the result would be to produce a surface horizon which is relatively richer in the coarser fractions than the parent material.

*Separation of minerals of different grades in the fine sand fraction**

The fine sand fractions were treated with oxalic acid to remove the cement-iron oxide material, washed, dried and then separated into three fractions, detailed below, by shaking with bromoform (sp. gr. 2·9) and a mixture of bromoform and benzene (sp. gr. 2·62):—

(a) Sp. gr. higher than 2·9, mostly ferro-magnesian minerals

(b) Sp. gr. between 2·62 and 2·9, mostly quartz grains

(c) Sp. gr. less than 2·62, mostly feldspar grains

These three fractions were washed, dried and separately weighed in chemical balances. This gave the percentages of different fractions in the fine sand fraction.

Table III shows the quantities of different fractions expressed as percentage of fine sand and of the soils.

The results show that the soils of Midnapur, Bengal, are the richest in the percentages of heavy minerals. The soils of Palamau (Daltonganj), Cuttack (Orissa), and of Puri (Orissa) are intermediate in their content of heavy minerals. The greater the percentages of heavy minerals present in the soil, the greater would be the probability that the soil is changing into a new one provided external disturbing factors like erosion be not present. These considerations, however, do not apply to the case when resistant minerals like garnet or iron oxide are present. On the basis of these general considerations we should expect that the soils obtained from Khasi hills, Assam, and also from the districts of Manbhum and Singhbhum have been weathered to a great extent, whilst the soil of Midnapur are comparatively immature.

*The silt fraction was not analysed quantitatively, because it was not found practicable to utilize the heavy liquid method of separation for material of so fine a grade.

The percentages of the middle and light fractions in the soil types do yield any useful information unless the percentages of minerals contain useful plant nutrients be separately determined.

TABLE III
Different fractions expressed as percentage of fine sand and of soil

Locality	Soil No.	Heavy fraction		Middle fraction		Light fraction		Error as per cent of fine sand
		Fine sand (per cent)	Soil (per cent)	Fine sand (per cent)	Soil (per cent)	Fine sand (per cent)	Soil (per cent)	
Hathwara Farm, Manbhum, Bihar	81p	1.2	0.252	2.1	0.440	90.4	19.0	6
	82p	1.8	0.315	1.7	0.298	88.4	15.5	8
	83p	2.2	0.475	2.2	0.475	88.4	19.1	7
	84p	2.2	0.26	2.4	0.284	85.2	10.1	10
	85p	0.5	0.1	3.3	0.66	89.2	17.84	7
	86p	7.8	1.87	37.9	6.5	48.8	8.35	5
Putida, Singhbhum, Bihar	87p	1.3	0.279	70.2	15.1	16.7	3.59	11
	88p	0.8	0.096	45.9	5.51	47.8	5.73	5
	89p	0.3	0.0465	60.2	9.33	32.6	5.05	6
Ratu, Ranchi, Bihar	90p	5.9	1.16	68.0	13.3	17.4	3.4	8
	91p	3.7	0.536	40.4	5.86	47.2	6.84	8
	92p	5.2	0.676	13.4	1.74	70.7	9.19	10
	93p	6.8	0.768	44.8	5.06	43.2	4.88	5
Daltonganj, Palamau, Bihar	94p	9.9	3.23	7.3	2.38	76.6	24.9	6
	95p	4.7	0.992	6.6	1.39	82.5	17.4	6
	96p	5.5	0.88	15.4	2.47	72.6	11.6	6
	97p	0.3	0.513	1.3	0.222	95.5	16.3	2
Tangi, Cuttuck, Orissa	98p	7.0	0.237	6.1	2.06	83.9	28.4	3
	99p	7.5	0.209	7.7	2.15	79.8	22.3	5
	100p	30.7	6.07	31.3	6.29	34.1	6.85	3
Dhanmandal, Cuttuck, Orissa	101p	7.6	1.81	11.9	2.83	76.7	18.3	3
	102p	10.8	0.972	7.1	0.639	77.4	6.66	4
Kapileswar, Bhubaneswar, Orissa	103p	9.2	1.93	37.7	7.91	49.2	10.3	3
	104p	12.8	2.48	33.9	6.58	51.5	10.0	1
	105p	5.4	0.848	14.9	2.33	77.6	12.2	2
Jhinkartangi, Khurda, Puri, Orissa	106p	9.3	0.948	32.6	3.33	54.1	5.52	4
	107p	12.8	1.41	36.2	3.98	44.8	4.94	6
	108p	16.3	1.91	20.6	3.04	58.2	6.81	4
	109p	25.1	6.24	45.2	11.25	26.5	6.60	3
	110p	2.4	4.92	19.6	4.90	74.6	15.3	3
Jalgarh, Midnapur, Bengal	112p	2.9	5.45	1.9	0.357	92.6	17.4	2
	113p	2.1	3.89	8.2	1.52	87.1	16.1	2
	114p	1.2	2.18	0.7	0.1274	96.5	17.6	1
	115p	0.9	0.374	0.8	0.332	95.6	39.7	2
	116p	2.1	9.38	5.2	2.32	91.1	40.7	1
Mailda, Midnapur, Bengal	117p	0.9	0.174	40.8	7.87	54.8	10.6	3
	118p	1.2	0.161	42.4	5.68	53.97	7.22	2

* Represents loss of material in working manipulation

TABLE III—*contd.*

Locality	Soil No.	Heavy fraction		Middle fraction		Light fraction		Error* as per cent of fine sand
		Fine sand (per cent)	Soil (per cent)	Fine sand (per cent)	Soil (per cent)	Fine sand (per cent)	Soil (per cent)	
Jhlang, Khasi Hills, Assam	119p	0.6	0.072	24.5	2.94	68.9	8.26	6.0
	120p	0.6	0.0678	13.9	1.57	78.5	8.87	7.0
	121p	0.8	0.0936	6.0	0.72	86.9	10.17	6.3
	122p	0.5	0.0475	3.7	0.352	86.7	8.24	9.1
Jorhat Hills, Upper Jorhat, Tura	144p	11.9	2.19			87.4	16.1	0.7
	145p	5.8	1.08			93.2	17.3	1.0
	146p	6.6	1.08			92.0	15.1	1.4
	147p	5.80	0.75			93.1	12.0	1.1

* Represents loss of material in working manipulation

In the case of these soils the data in the column of light fraction represent both the light and middle fraction

It seems on the whole that the soils generally are fairly rich in their spar constituents. The proportions of the middle fraction which consist mainly of quartz particles are, however, not so high except in a few cases (e.g. Jhlang, Ranchi and Midnapur).

Determination of minerals in the heavy fraction of the fine sand

The separated fine sand fractions were mounted on microscopic slides with Canada balsam by following essentially the procedure described by Fisher [1932]. It was found that the nature of the minerals in the middle and in the light fractions of all the soil samples were very nearly the same. The mineralogical composition of these fractions was generally as follows: mainly quartz and felspar grains and some kaolinized feldspars; most of the quartz and felspar grains were found to be stained with limonitic materials; limonitic minerals were also found.

Detailed quantitative mineralogical studies were undertaken with the heavy fractions only.

Determination of different minerals in the heavy fractions

A square-type graduated micrometer scale was used inside the microscope eye-piece. The microscope tube was adjusted so that the micrometer scale was in the field of view along with the minerals of the microscopic slide and the total number of minerals in the graduated square area were counted. The number of individual minerals in the same area were also determined. By a simple process of calculation the percentages by volume of the various minerals in the microscopic slide could be calculated and multiplying these volume percentages by the specific gravity of the minerals, the percentages by weight of the minerals can be obtained. It is, however, necessary to take a large number of counts in order that the calculated percentages by weight of the minerals might be approximately correct. If a large number of counts are taken, it is found that the percentages of the various minerals agree within 10 per cent.

Table IV gives in one place the data on the percentages by weight of various minerals in the heavy fraction of the fine sand along with the nature of the parent materials of the soil types.

Weight of minerals in

Locality	Soil No.	Depth	Iron oxide**	Zircon	Epidote	Horn-blende	Tourmaline
Hathwara, Manbhumi, Bihar	81p	0—1 ft. 6 in.	67.5	109.0	24.2	35.3	
	82p	1 ft. 6 in.—2 ft. 3 in.	85.5	171.0	19.7	34.8	
	83p	2 ft. 3 in.—3 ft. 6 in.	147.0	232.0	36.0	40.5	5
	84p	3 ft. 6 in.—4 ft. 11 in.	79.5	108.0	22.2	15.7	7
	85p	Below 4 ft. 11 in.	35.5	36.2	13.4	8.85	1
	86p	Below 30 ft.	93.5	1150.0	23.5		
Putida, Singhbhum, Bihar	87p	0—1 ft.			74.0	5.44	
	88p	1 ft.—2 ft. 9 in.	4.34	4.2	3.11		
	89p	2 ft. 9 in.—4 ft.	2.59		0.77		
Ratu, Ranchi, Bihar	90p	0—1 ft.	1050	27.6	30.6	54.0	
	91p	1 ft.—2 ft.	460	5.36	22.3		3
	92p	2 ft.—3 ft.	624	8.12	24.0	21.2	
	93p	3 ft.—4 ft.	715	9.24	13.7	24.1	6
Baralota, Daltonganj, Bihar	94p	0—1 ft. 11 in.	514			2540.0	36
	95p	1 ft. 11 in.—2 ft. 9 in.	302		6.75	675.0	
	96p	2 ft. 9 in.—4 ft.	239			573.0	
	97p	4 ft.—6 ft.	134	217.0		84.6	
Tangi, Orissa	98p	0—1 ft.	30.6		88.5		
	99p	1 ft.—2 ft.	25.1		86.0		
	100p	2 ft.—4 ft.	16.6		39.8		
Dhanmandal, Cuttack, Orissa	101p	0—5 in.	437		360		
	102p	5 in.—4 ft.	333		204		
Kapileswar, Bhubaneswar, Orissa	103p	0—2 ft. 11 in.	1800	47.5	35.1		
	104p	2 ft. 11 in.—4 ft.	2400		44.8		
	105p	Below 30 ft.	675		71.5		
Jhinkartangi, Khurda town, Orissa	106p	0—1 ft.	687	180	66.5		
	107p	1 ft.—2 ft.	935	407	60.2		
	108p	2 ft.—3 ft. 6 in.	1220	494	83.0		
	109p	3 ft. 6 in.—10 ft.	4330	1270	111.0		
	110p	30 ft.—50 ft.	4120	415	307.0		
Lalgah, Midnapore, Bengal	112p	0—4 in.	4000	205	353		578
	113p	4 in.—3 ft. 4 in.	3090	96	71	62.6	282
	114p	3 ft. 4 in.—4 ft.	1470	162	120		211
	115p	7 ft.—8 ft.	273	18.7	24.1		36.6
Malda, Midnapore, Bengal	116p	0—8 in.	6030	463.0	770.0	453	906.0
	117p	Bed soil of Cossye R.	199.5	8.75	14.5		19.9
	118p	48 ft. below	112.0	8.30	16.9		18.9
Mawphlang, Khasi Hills, Assam	119p	0—6 in.	53.8	2.71	9.35		1.7
	120p	6 in.—1 ft. 3 in.	49.4	4.93	7.38		1.06
	121p	1 ft. 3 in.—2 ft. 1 in.	62.0	4.88	12.30		3.11
	122p	2 ft. 1 in.—4 ft.	31.4	2.38	4.40		3.8
Upper Chandmar, Tura, Garo Hills, Assam	144p	0—3 in.	1930	184.0	77.5		
	145p	3 in.—1 ft. 8 in.	900	104.0	76.7		
	146p	1 ft. 8 in.—2 ft. 8 in.	925	104.0	1.93		
	147p	2 ft. 8 in.—4 ft.	643	10.6			

*Calculated from the percentages of heat

** These represent resistant forms of oxides of iron white

r 10⁵ gm. of soil*

Ortho- quartzite	Garnet	Augite	Chlo- rite	Cal- cite	Dolo- mite	Rutile	Bio- tite	Kya- nite	Parent material
	4.5 2.76 6.41 4.95	8.05 7.65 8.88 3.34	3.29 2.02 4.68 1.45 1.37						Granite and syenite
							75.7		Do.
						72.5 843.0 414.0		126.5 6.15 1.7	Dalma traps and Mergui volcanics
13.2									Unclassified crys- talline gneiss, etc.
	23.3 7.54 13.7 72.0	41.7 12.2	50.9 13.1 25.0			19.0 6.15 16.7		5.89	Limestone, shales and slates
						118 98 545			Older alluvium and laterite
						1020 435			Laterite mixed with granite and sandstone
		23.8	78.0					44.0 28.1	Gniess Do.
	6.74				14.0				Do.
			45.3	88.5	69.8 329.0 74.0	41.4 69.0			Do.
74 54 52 15.0			2.82		42.6 89.7 32.8 2.91	44.2 24.9			Older alluvium and laterite Do.
58.0 3.52 5.0		1.61	140.0 1.32		72.2 2.72 1.30		3.83	1.47	Older alluvium and laterite Do. Do.
			1.63 2.01 7.90 0.72		1.69 1.48 1.49	0.83 3.06 2.19 3.29			Shillong series
			3.16						Fab-sandstone

mineral in the soil (Table III)

could not be removed by treatment with oxalic acid

On the basis of the data given in Table IV, the soil profiles studied may be tentatively grouped as follows :—

1. Zircon preponderant . . . (a) Hathwara Farm (Purulia, Bihar)
(b) Jhinkartangi (Khurda Town, Orissa)
(c) Upper Chandmari (Tura, Garo Hills, Assam)
2. Hornblende preponderant . . . Baralota (Daltonganj, Bihar)
3. Rutile preponderant . . . (a) Putida (Singbhum, Bihar)
(b) Tangi (Cuttack, Orissa)
(c) Dhanmandal (Cuttack, Orissa)
4. Epidote preponderant . . . (a) Tangi (Cuttack, Orissa)
(b) Dhanmandal (Cuttack, Orissa)
(c) Mawphlang (Khasi Hills, Assam)
5. Tourmaline preponderant . . . Midnapore (Bengal).

Mention may be made here of the work of Jeffries [1937], who working with some Pennsylvania soils, has concluded that the percentages of minerals present in the soils can be used as an important aid in soil classification.

SUMMARY AND CONCLUSIONS

The present work was undertaken with a view to characterizing some profiles of red and lateritic soils collected from different parts of India by finding out the mineralogical composition of the fine sand fractions of the profile samples, and from the mechanical composition of the soils and the chemical composition of the clay fractions.

In the initial stages of the mineralogical work, the fine sand fractions with which the microscopic determinations were carried out, have not been estimated quantitatively for the percentages of heavy, middle and light fractions. At this stage of the work the heavy and light fractions were separated by shaking with bromoform (sp. gr. 2.9) only, but the separated portions were not weighed. Also the nature of the minerals on the microscopic slides was only qualitatively determined, but they were not counted for determining the percentages of different minerals in the slides.

In the later stage of the microscopical work it was felt desirable to obtain the mineralogical data of the fine sand fractions on a quantitative basis. The fine sand fractions were accordingly separated into three fractions, viz. heavy (sp. gr. 2.9), middle (sp. gr. 2.9-2.65), and light (sp. gr. 2.65) which were weighed separately and their mineralogical compositions determined with the help of a petrological microscope. The percentages of different minerals present in the heavy fractions were also carefully counted with the help of a graduated cross-wire eye-piece micrometer scale.

Common ferro-magnesian rock-forming minerals are represented by one or more species in all the soils examined, one or the other being present in preponderating amount. Hornblende was occasionally present. Chlorite was found in most of the soils studied. Biotite was occasionally found (e.g. Nilgiri hills). These minerals are of resistant nature and they are concentrated in material which has been subjected to prolonged weathering and has accumulated from a variety of sources. Epidote and garnet were frequently found in granular form, whilst tourmaline and especially zircon, though sometimes fragmentary, showed crystalline forms.

Iron minerals like haematite, magnetite and limonite were present in all the soils examined.

With regard to the percentage of the ferro-silicate group of minerals, the soils of basic igneous origin are naturally the richest.

In almost all the cases the proportion of rarer accessory minerals like zircon, garnet and tourmaline is higher compared with the common -forming minerals like biotite and hornblende. The latter minerals are potential sources of plant food supply and are usually present in the soil in small amounts, which explains the comparatively little unproductivity of the soil types.

The general nature and proportions of minerals in the light (sp. gr. less than 2.65) and middle fractions (sp. gr. between 2.9 and 2.65) of all soil samples were found to be very similar. In the light fraction the assemblage of minerals consisted mostly of feldspar and grains of quartz often stained with oxides of iron. In the middle fraction, on the other hand, the predominating minerals were quartz grains, mixed with some feldspars, both being stained with oxides of iron.

The outstanding minerals present in the heavy fraction are iron oxide, zircon, tourmaline, staurolite, chlorite, hornblende, epidote and rutile. In a few cases, the existence of minerals like leucocene, augite, dolomite, calcite and biotite were noticeable.

On the basis of the percentages of the mineralogical constituents in the soil samples, the soil profiles have been tentatively grouped into five classes.

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DETERMINATION OF ORGANIC PHOSPHORUS IN ALKALI EXTRACTS OF SOILS

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DEAN [1938] outlined a colorimetric method for determining inorganic organic phosphorus in the alkali extracts of soils. The colour of the extract was removed either by using kieselguhr as an absorbent or by oxidizing it by means of sodium hypobromite at 95°—100° C. The amount of phosphorus determined colorimetrically on the decolourized extract was assumed to be inorganic and the difference of the inorganic phosphorus thus determined and the total phosphorus to the extract determined separately by ashing was taken to represent the organic phosphorus fraction of the extract. The principle underlying this separation is that the organic phosphorus compounds do not liberate any free phosphate ion by the decolourizing treatments. The high temperature at which bromine oxidation was carried out by Dean suggests the possibility of some organic phosphorus being decomposed into inorganic form in which case the separation would become absolutely untrustworthy. Moreover, to get reproducible results it is essentially necessary that the conditions of bromine oxidation should be clearly specified. The work was undertaken with a view to examining the conditions of bromine oxidation, in obtaining a method which would allow the organic phosphorus matter to be removed without destroying any organic phosphorus compounds. For this purpose the alkali extracts of five soils (A. E. A. samples) were decolourized by various treatments as shown below.

DECOLOURIZING TREATMENTS

(1) Bromine oxidation at boiling temperature, excess bromine being boiled off after acidification (Dean's procedure).

An aliquot of the alkali extract was treated with 5 c.c. of bromine water and heated on a water bath until decolourization was complete. The extract was then acidified to liberate the excess bromine. The bromine was finally boiled off on a water bath until the extract was colourless.

(2) Bromine oxidation at 40°C., excess bromine being boiled off.

The brominated extract was heated on a water bath which was kept at 40°C. The excess bromine was boiled off as in (1).

(3) Bromine oxidation at 40°C., excess bromine being removed at 40°C.

The bromine oxidation was done at 40°C. as in (2). The extract was acidified as before and then heated on a water bath kept at about 40°C. until all the bromine was driven off.

(4) Bromine oxidation at room temperature, excess bromine being removed by aeration.

Bromine water was added to the extract and the extract allowed to stand for some time. It was then acidified as before and a current of air bubbled through it until it was colourless.

(5) Kiesselguhr method (Dean's procedure)—An aliquot of the extract was acidified and heated. The coagulated humus was filtered off and the filtrate shaken with kiesselguhr and again filtered.

(6) Kiesselguhr method (modified)—The humus was coagulated as in (5) but not filtered off before shaking with kiesselguhr.

The inorganic phosphorus was determined in the colourless extract after decolourization had been effected by the above treatments. The total phosphorus of the original alkali extract was estimated by ashing (as shown later) and the organic phosphorus was obtained by difference. The colorimetric method of Deniges [1920] as improved by Truog and Meyer [1929] was used all through. The results showing the organic phosphorus figures only are given in Table I.

TABLE I

Effect of bromine oxidation at different temperatures on the organic phosphorus in the alkali extract

(Organic P_2O_5 in mg. per 100 gm. of soil)

Soil	A 1441 Broadbalk head land, heavy loam, acid	A 1442 Woburn, sandy loam, acid	A 1443 Bangor, loam, neutral	A 3328 Carbello, loam, acid	A 2865 King's lynn, sandy loam, alkaline
Treatments					
1. Oxidation and expulsion of bromine at 100°C.	46	24	132	45	23
2. Oxidation at 40°C. and expulsion at 100°C.	48	..	130
3. Oxidation and expulsion at 40°C.	62	32	156	52	32
4. Oxidation and expulsion at room temperature	64	33	160	56	32
5. Kiesselguhr (Dean's)	64	33	160	56	35
6. Kiesselguhr (modified)	65	33	160	56	33
Total P_2O_5 extracted by al- kali	126	74	320	96	72

It will be seen from the table that the temperature of bromine oxidation has a great deal of effect on the organic phosphorus determined. If both

bromination and expulsion of excess bromine are carried out at 100°C. as in (1), the organic phosphorus figures are much lower as compared with the figures obtained by the Kiesselguhr method which shows that at this high temperature the organic phosphorus compounds are considerably broken down by bromine, to be determined as inorganic phosphorus. It has, of course, been assumed that no such transformation takes place by the treatment with kiesselguhr. When both the operations are carried out at 40°C. as in (3), the results are comparable with those of the Kiesselguhr method. But again if only the expulsion is done at 100°C. as in (2), the figures fall down to the original values as obtained in (1), from which it becomes evident that neither the bromine oxidation nor the expulsion of the excess bromine can be undertaken at 100°C. without affecting the organic phosphorus fraction. Though the results are not affected by the treatments in (3), the removal of excess bromine at 40°C. was found to take more than 2½ hours. Bromination at room temperature and removal of excess bromine by aeration as in (4) compare very favourably with the kiesselguhr and the low temperature oxidation methods. The cold bromination also simplifies the procedure to a great extent, the reaction is completed in a few minutes and the excess bromine can also be removed in a short time. This, therefore, affords a simple and efficient means for the separation of the inorganic and organic phosphorus in the alkali extracts of soils.

As regards the kiesselguhr method it will be seen that the procedure can be a little simplified as in (6) without affecting the results. This method is not only long and tedious but is also incapable of general application. Kiesselguhr can be used as a decolourizing agent only in the case of lightly coloured extracts. Experience showed that even moderately coloured extracts could not be decolourized by it.

Subsequent experience with a large number of determinations on a variety of soils showed that the cold bromination method can be applied successfully to all soils. An occasional highly organic soil may require large amounts of bromine water. While working with some black fen soils giving highly coloured alkali extracts, the author had the opportunity to compare again the original method of Dean with the method proposed by him. Table II contains the results of organic phosphorus determination by the two methods in the successive alkali extracts of two fen soils. Kiesselguhr method was completely ineffective in these cases.

The proposed method gives higher values of organic phosphorus in all the extracts of the two soils. This conclusively shows that the organic phosphorus compounds are considerably decomposed during the process of bromine oxidation at 100°C. as originally proposed by Dean. This breakdown is not, however, proportional to the total organic phosphorus in the extract. It probably depends upon the nature and kind of organic phosphorus compounds and therefore varies from soil to soil. It will be noted that in the second extract of soil A 3560-1 the whole of the organic phosphorus was broken down.

The method finally proposed for the determination of inorganic and organic phosphorus in the alkali extracts of soils may be stated as follows:—

An aliquot of the alkali extract (however obtained) is measured into a 100 c.c. conical flask and treated with a saturated solution of bromine in water,

the volume of bromine required varying from 2 to 15 c.c. according to the colour of the extract. When a large amount of bromine is used, an equivalent amount of alkali should be added if the extract is not sufficiently alkaline already. The brominated extract is allowed to stand for a quarter of an hour and is then acidified with 6N sulphuric acid until bromine is liberated. The flask is then connected to a filter pump and a current of air is bubbled through to remove free bromine. When the extract is colourless it is washed into a measuring flask (50 or 100 c.c.), made up to volume, filtered and the phosphorus determined colorimetrically on an aliquot.

TABLE II

Comparison of the original method of Dean and the proposed method of determining the organic phosphorus in alkali extracts

(Organic P_2O_5 expressed as mg. per 100 gm. of soil)

Soil	Method	1st extract	2nd extract	3rd extract	Total
3560-1 Littleport, heavy neutral fen	Original method	25.0	0.0	4.0	29.0
	Proposed method	54.0	17.0	5.2	76.2
3246-47 Peterboro, heavy acid fen	Original method	55.0	10.0	5.8	70.8
	Proposed method	80.0	17.0	8.7	105.7

The total alkali soluble phosphorus is determined on an aliquot which is evaporated to dryness with 2 c.c. of 10 per cent solution of magnesium nitrate in a porcelain basin and gently ignited. The ignited residue is treated with 1 c.c. of conc. hydrochloric acid, the basin being covered with a watch glass. It is then diluted with water to about 20 c.c. and heated on a sand-bath for 15 minutes. The extract is made to a known volume and phosphorus determined on an aliquot. The organic phosphorus is obtained by difference.

SUMMARY

1. The colorimetric method for the estimation of inorganic and organic phosphorus in alkali extracts of soils as proposed by Dean has been critically examined.
2. It has been found that the bromine oxidation at 100°C. breaks down a part of the organic phosphorus compounds.
3. On the other hand, the oxidation at room temperature does not decompose any organic phosphorus.
4. A simplified procedure for the separation of organic and inorganic phosphorus in the alkali extracts of soils by bromine has been described.

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SEEDLING-ADULT COLOUR RELATIONSHIPS AND INHERITANCE IN SORGHUM

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(With Plate IX)

PLANT pigments can be classified broadly into two groups, the vegetative and the ornamental. The vegetative pigments are those that are related to the chlorophyll and are therefore indispensable for the normal metabolic activities of the plant. The ornamental pigments have a comparatively less important rôle and though several functions of a physiological and biological nature are attributed to them, they do not seem to be vital to plant life. Sorghum seedlings manifest various types of pigmentation soon after they emerge from the soil and after a few days of growth. Several colour patterns that are associated with chlorophyll exist in sorghum. Under the term 'seedling colours' referred to in this paper are included the colours other than those that are related to chlorophyll. These are confined either to the cell-sap or to the cell-wall and do not exist in the plastid condition like those of the chlorophyll group.

Attempts to classify the sorghum seedlings into pigmentation types have been made in the past. Reed [1930] classified them into two groups, those with red-coloured coleoptiles and those with green coleoptiles. The seedlings with red coleoptiles proved a simple dominant to those with green coleoptiles. Rangaswami Ayyangar [1930, 1932] recorded a simple monogenic segregation between purple and green seedlings. Rangaswami Ayyangar [1934], Rangaswami Ayyangar, Ponnayya and Reddy [1938] refer to the segregation of purple and green coleoptiles. Karper and Conner [1931] made mention of red-stemmed seedlings which behaved as a simple dominant to non-red stemmed seedlings. Fevorow and Havenselman [1934], investigating the hybrids of sorghum and sudan grass, mentioned that the violet colour of the shoot of sudan grass is due to the presence of one dominant factor. Woodworth [1936] reported a 9 : 7 segregation between red-stemmed and green-stemmed seedlings in a cross between Shallu and Black Spanish; Rangaswami Ayyangar, Rao and Reddy [1938] have reported a simple segregation of deep purple and purple plumules close on emergence from the soil. The deep purple pigment of the plumules always went with the purple colour of the anthers.

This pigmentation in sorghum seedlings was made use of by plant breeders in several instances. It is an old observation at the Millets Breeding Station,

Coimbatore (south India), that in pure breeding lines of sorghum, odd seedlings possessing a different seedling colour from the type occur very often. These seedlings later on prove to be the resultants of natural crossing. If allowed to grow to maturity they contaminate the inbreeding line still further as sorghum is easily susceptible to cross-pollination and up to as much as 50 per cent of natural crossing is reported in adjacent rows [Ball, 1919]. Hence these seedlings are pulled out in order to prevent further contamination of the inbreeding line. Reed [1930] was able to detect sorghum hybrids at the seedling stage itself with the help of the dominant seedling colour. He selected a parent possessing the recessive seedling colour as mother and pollinated it with a male parent having the dominant seedling colour. The mother parent was not emasculated before pollination. When the seeds of this parent were sown, both the mother type and hybrid seedlings were produced. The hybrid seedlings were readily distinguished from the mother type by the help of the dominant seedling colour which they possessed. Rangaawami Ayyangar [1930] made use of the segregation of purple and green seedlings for demonstrating a simple mendelian segregation graphically by germinating and growing the seedlings on the earhead itself. Vinall, Stephens and Martin [1936] distinguished red and green coleoptiles and made use of this colour for the classification and identification of sorghum varieties of the United States of America.

During the years 1930-36, a large number of sorghum varieties from several important sorghum centres of the world were obtained and grown at the Millets Breeding Station, Coimbatore. Among these, many varieties, especially those from Africa, exhibited certain types of seedling colours which have not so far been recorded. In a previous paper [Rangaawami Ayyangar, 1934] mention has been made of sorghum seedlings with coloured and colourless roots. This aspect was explored and elaborated further; this has led to the knowledge of the existence of several types of root pigmentation. Previous investigators classified sorghum seedlings according to the nature of the shoot pigmentation. The present classification is made based on both the shoot and root pigmentations.

CLASSIFICATION OF SORGHUM SEEDLINGS BASED ON PIGMENTATION

SHOOT PIGMENTATIONS

The plumules (the seedling shoots) of sorghum seedlings soon after their emergence from the soil are either purple or green. When they are coloured purple they may be either deep purple or purple. The shoot pigmentations are therefore classified into three groups, (1) deep purple, (2) purple, and (3) green. Detailed descriptions of these three groups are given below.

Deep purple shoots

The coleoptile which is the first to emerge from the soil rapidly develops a deep purple tint (Amaranth purple to Aster purple of plate XII in Ridgway's [1912] *Colour Standards*). The young shoot which pushes out of the coleoptile is pigmented likewise. The first two leaves are coloured deeply like the coleoptile. But in later leaves the pigment gradually lightens up and altogether disappears from the sixth to the eighth seedling leaf upwards. As the shoot grows and when the leaf blades begin to spread out, the pigment disappears

from the blade area and is confined to the region of the leaf-sheath only. In seedlings about a week to ten days old and with about four to five leaves, the pigment is seen only on leaf-sheaths. This pigment on leaf-sheaths is very conspicuous and extends even to the bases of the older leaf-sheaths which are at or near the ground level, so much so that even in seedlings about 40 days old the shoot pigment is easily distinguished.

Besides the manifestation of the pigment on the sheath and the blade areas, the auricular junctions (the specialized tissue that connects the blade with the sheath) also get coloured purple from the third leaf onwards. In sorghum, the junctions of the first two leaves are not well developed and the pigment is not seen in these regions. From the third leaf onwards they get well elaborated [Rangaswami Ayyangar, Rao and Rajabhooshanam, 1938] and the pigment is very clearly noticed. Seedlings over a week to ten days old manifest this pigment well. In seedlings of about 20 days old the exposed portions of the outer margins of the leaf-sheaths also develop purple pigment. The development of the purple pigment in the auricular junctions and the leaf-sheath margins is a characteristic of this class of seedlings and these pigmented organs are unmistakable guides in distinguishing this type of shoot pigmentation from the rest.

Purple shoots

In this type the manifestation of the pigment is almost parallel to that of the deep purple type, except that the colour is of a much lighter shade of purple (pale Rosolane purple to light Rosolane purple of plate XXVI in Ridgway's [1912] *Colour Standards*). The coleoptile when emerging from the soil is of a pale greenish tint but very soon becomes purple when outside the soil. The plumular leaves as they come out from the coleoptile are also coloured purple. This colour of the plumular leaves is always of a lighter tint than that of the coleoptile. Except the first two seedling leaves the older leaves do not manifest the purple pigment in the blade area. Even in these two seedling leaves the pigment disappears from the leaf-blade, when they have fully emerged from the coleoptile, and is confined to the leaf-sheaths. Similar to the deep purple shoots, in this type also the bases of the leaf-sheaths at or near the ground level are pigmented purple in seedlings of about 40 days old, though in a faint measure. The auricular junctions and the leaf-sheath margins do not develop purple pigment in this type but remain yellowish green. Besides the difference in the depth of the pigment in the young shoots, the non-manifestation of the pigment in leaf-junctions and leaf-sheath margins clearly helps to distinguish this type of shoot pigmentation from the deep purple type.

Green shoots

In this type the coleoptile and the plumular leaves do not develop purple pigment at any stage of the seedling. The coleoptile when emerging from the soil is either pale yellow or yellowish green. The plumular leaves that push out of the coleoptile are green and remain so thereafter without any trace of purple pigment. The basal region of the older seedling leaves which are at or near the ground level are either yellowish green or green. The auricular junctions and the leaf-sheath margins remain unpigmented. This type

is easily distinguished from the other two pigmented types by the absence of the purple pigment in the coleoptile, plumular leaves, auricular leaf-junctions and leaf-sheath margins.

ROOT PIGMENTATIONS

In sorghum as in other mono-cotyledonous plants the primary root soon dies away and a very large number of secondary roots develop, chiefly from the base of the coleoptile. These roots form the mainstay for the growth of the plant. In young seedlings (up to two to three weeks old) the roots are either colourless or occasionally have a light straw colour. But in seedlings over 20 days old they begin to develop their respective colours. They may be either coloured purple or brown. When coloured purple they may be either reddish purple or blackish purple. There are therefore three pigmented groups in roots, viz. (1) Reddish purple, (2) Blackish purple, and (3) Brown. These three groups are described below:—

Reddish purple roots

These roots at first sight appear brick-red, but on closer examination a faint tint of purple is revealed (Coral-red to Eugenia-red of plate XIII in Ridgway's [1912] *Colour Standards*). The pigment develops first in patches on the older roots in seedlings over 20 days old. These patches gradually begin to coalesce with one another till the entire root looks pigmented. The pigment is not uniformly distributed. On the same root deeper and lighter coloured patches appear. In the early stages of manifestation the pigment is of a lighter tint and gradually deepens later. Generally the older roots get pigmented first, but there is no regularity in colour development on the individual roots. This pigment persists throughout the life of the plant and even after its death in dead tissues.

Blackish purple roots

In this group the roots are coloured black with a slight tinge of purple (Prune purple to Blackish purple of plate IX in Ridgway's [1912] *Colour Standards*). The development, the manifestation and the persistency of the pigment are similar to those of the reddish purple root pigment. The distinction between the reddish purple and the blackish purple colour is clear enough in freshly washed roots. The difference is more markedly seen when the roots are washed and dried in the sun. The reddish purple roots become brick-red in colour, while the blackish purple roots turn black. On drying, the purple part of the pigment which is common to both these types seems to become unobtrusive, the reddish and the blackish nature of the pigment getting emphasized.

Brown roots

These roots do not develop purple pigment, but are coloured yellowish brown to brown (Cream buff to Chamois of plate XXX in Ridgway's [1912] *Colour Standards*). The development, the manifestation and the persistency of the pigment are exactly like those of the purple group. This type is easily distinguished from the other two in that the roots do not possess purple colour at any stage of the plant. When the roots are washed free from the soil, the moist roots are coloured yellowish brown. On drying, this yellow colour disappears and the roots look mere brownish.

DIFFERENT TYPES OF SEEDLING COLOURS

In sorghum seedlings three kinds of shoot pigmentations and three kinds of root pigmentations were described in the previous pages. These shoot and root pigmentations are two quite different sets of colour manifestations. Any one of the three shoot pigmentations can combine with any one of the three root pigmentations. This combination results in nine different types of seedling colours. These are given below.

No.	Seedlings		Reference to Plate IX
	Shoot colour	Root colour	
1	Deep purple . . .	Reddish purple . . .	Fig. 1. A-D
2	" . . .	Blackish purple . . .	" 4. A-D
3	" . . .	Brown . . .	" 7. A-D
4	Purple . . .	Reddish purple . . .	" 2. A-D
5	" . . .	Blackish purple . . .	" 5. A-D
6	" . . .	Brown . . .	" 8. A-D
7	Green . . .	Reddish purple . . .	" 3. A-D
8	" . . .	Blackish purple . . .	" 6. A-D
9	" . . .	Brown . . .	" 9. A-D

RELATIONSHIP OF SEEDLING COLOURS TO ADULT PLANT PIGMENTATIONS

The sorghum plant manifests purple pigmentation in various places and at various stages in its life period, and the inheritance of these pigments have been studied by several investigators. The relationship of the seedling colours to the adult plant pigmentation and some of these pigmentations are discussed below.

The shoot pigmentation of the seedling were found to have an absolute relationship with the pigment of the leaf-axils—the basal portion of the inside of the leaf-sheath above the region of the nodal band, for brevity called the nodal band [Rangaswami Ayyangar, Rao and Reddy, 1938]. When the shoots are green the axils are green. When the shoots are deep purple the axils of the plant are likewise coloured deep purple. In addition, the nodal bands, the auricular leaf-junctions and the outer margins of the leaf-sheaths extending from the auricular junctions above to the nodal band below, are also coloured purple. The manifestation of the purple pigmentation in these regions is at its best at the time when the plants are in flower immediately after flowering. This close association in colour manifestation between the seedling and the adult has been so constant that not even an exception was met with in the several thousands of pure lines and numerous segregating families studied. Hence it is presumed that the pigmentation in these places is being brought about by the play of the same set of genes. The close relationship between the deep purple pigmentation of the plumules on emergence from the soil (the seedling shoots), the axils, the nodal bands, the auricular junctions, the leaf-sheath margins and purple colour of the flowers was reported by Rangaswami Ayyangar, Rao and Reddy [1938]. In this type of deep purple pigmented shoots the anthers may or may not be coloured purple. When the shoots are deep purple in colour, irrespective

of the fact whether the anthers are purple or not, the axils, the nodal leaf-junctions, and the leaf-sheath margins are always coloured purple.

The root colours have an absolute relationship with those of the shoot. The glume pigmentations reported by Rangaswami Ayyangar [1933]. When the roots are coloured purple, the leaf-sheaths and glumes are also coloured purple. When the roots are brown, the leaf-sheaths and glumes are brown. In the purple group, the reddish purple roots are associated with reddish purple leaf-sheaths and glumes and the blackish purple roots with blackish purple leaf-sheaths and glumes. Beside this concurrent manifestation of the pigmentation on the root, leaf-sheath and glume, even portions of midrib, leaf-blade and leaf-sheath that get injured when in active growth develop the corresponding root colour. The pedicelled spikelets, the pulvinor regions of the panicle branches and the stylar end of the grain, in some cases, develop this root pigmentation.

The nine types of seedling pigmentations and their relationship with adult plant pigmentations presented are in the tabular statement below.

No.	Seedling pigmentation			Adult pigmentation	
	Shoot	Root	Axil	Leaf-junctions, leaf-sheath margins and nodal bands	Leaf-sheath and glumes
1	Deep purple	Reddish purple	Deep purple	Purple	Reddish purple
2	"	Blackish purple	"	"	Blackish purple
3	"	Brown	"	"	Brown
4	Purple	Reddish purple	Purple	Green	Reddish purple
5	"	Blackish purple	"	"	Blackish purple
6	"	Brown	"	"	Brown
7	Green	Reddish purple	Green	"	Reddish purple
8	"	Blackish purple	"	"	Blackish purple
9	"	Brown	"	"	Brown

GENETIC ANALYSIS OF THE SEEDLING COLOURS

Shoot pigmentations

The inheritance of the shoot pigmentation was pursued in 20 crosses. In these the inheritance up to the third generation was studied in eight crosses. The results are discussed below:—

A factor **PC** [Rangaswami Ayyangar, 1938] is responsible for the presence of the purple colour in the coleoptile and the young shoot. In its absence (**pc**) the coleoptile and the shoot are green. Purple (**PC**) is a simple dominant to green (**pc**). A factor **PJ** operating in the presence of the purple factor **PC** makes the coleoptile and the shoot deep purple and produces purple pigmentation in the leaf-junctions and leaf-sheath margins in addition. **PJ** has no visible effect in the absence of **PC**. These factor pairs **PCPC** and **PJPJ** result in the following genic constitution of the three groups:—

1. Deep purple **PCPCPJPJ**
 2. Purple **PCPCpj pj**
 3. Green **pcpcPJPJ**
- or **pcpcpj pj**

The interplay of these two factors results in the usual monohybrid and dihybrid ratios. Segregations for these three shoot pigmentations are presented in the following tables. Tables I—III give simple monogenic segregation and Table IV gives the dihybrid segregation.

TABLE I

Pure for pj and segregating for PC

(Shoots purple to green)

Segregation	Family number	Character of selection	Behaviour of progeny	
		Shoot colour	Shoot colour	
			Purple	Green
Monohybrid	A S 2177	Purple		
	" 1633	Green		
	" CLIV	Purple		
	" 3717	202	68
	" 3718	375	136
	" 3719	152	61
	" 3720	340	99
		Total	1069	364
		Expected 3 : 1 ratio	1074.75	358.25
		$\chi^2 = 0.120$	$P > 0.70$	
	A S 4456	Purple	186	78
Monohybrid	" 4457	"	650	237
	" 4460	"	593	207
	" 4461	"	361	109
	" 4462	"	485	115
	" 4463	"	324	120
	" 4465	"	63	20
	" 4466	"	679	216
	" 4470	"	187	69
	" 4459	"	Pure	..
	" 4468	"	"	..
	" 4469	"	"	..
	" 4465	Green	..	Pure
	" 4458	"	..	"
	" 4464	"	..	"
	" 4467	"	..	"
		Total	3528	1171
		Expected 3 : 1 ratio	3524.25	1174.75
		$\chi^2 = 0.0159$	$P > 0.80$	

The inheritance of this factor pair **PCPC** was pursued in 40 other families. These gave a total of 6815 seedlings with purple and 2238 seedlings with green shoots in the F_2 generation and 6071 purple and 1958 green in the F_3 generation.

TABLE II
Pure for PJ and segregating for PC
(Shoots deep purple to green)

Generation	Family No.	Character of selection	Behaviour of progeny	
		Shoot colour	Shoot colour	
			Deep purple	Green
F_1	A S 3999	Deep purple		
F_2	" 4847	"	369	12
	" 4848	"	252	8
	" 4849	"	291	10
		Total	912	31
		Expected 3 : 1 ratio	922.5	30
		$\chi^2 = 0.4780$	$P > 0.30$	
F_3	A S 5725	Deep purple	284	10
	" 5726	"	275	10
	" 5727	"	160	6
	" 5729	"	431	14
	" 5731	"	204	7
	" 5732	"	240	8
	" 5728	"	Pure	8
	" 5730	"	"	8
	" 5733	Green	"	Pur
	" 5734	"	"	"
		Total	1594	56
		Expected 3 : 1 ratio	1617	53
		$\chi^2 = 1.3085$	$P > 0.20$	

The same experience was met with in eight other families and the segregation figures of the F_2 generation are 375 deep purple and 110 green and in the F_3 generation 2547 deep purple and 840 green shoots.

TABLE III
Pure for PC and segregating for PJ
 (Shoots deep purple to purple)

Generation	Family No.	Character of selection		Behaviour of progeny	
		Shoot colour		Shoot colour	
				Deep purple	Purple
Parents	A S 3447 .	Deep purple			
	„ 60	Purple			
F ₁	„ CCXV	Deep purple . .			
F ₂	„ 4756 .	„ . .		474	167
	„ 4757 .	„ . .		366	127
		Total .		840	294
		Expected 3 : 1 ratio		850.5	283.5
				$\chi^2 = 0.5185 \quad P > 0.30$	
F ₃	A S 5511 .	Deep purple . .		235	85
	„ 5512 .	„ . .		210	65
	„ 5513 .	„ . .		204	67
	„ 5514 .	„ . .		Pure	67
		Total .		649	217
		Expected 3 : 1 ratio .		649.5	216.5
				$\chi^2 = 0.00154 \quad P > 0.95$	

The above experience was met with in 27 other families. These gave a total of 1386 deep purple and 462 purple in the F₂ and 6376 deep purple and 2029 purple shoots in the F₃ generations.

The dihybrid segregation involving both the factors **PC** and **PJ** was pursued in a family M S 1428 for three generations. M S 1428 is a selection from Tanganyika with purple shoot colour. In it a natural cross A S 3949 with deep purple shoot colour was obtained. When sown, this segregated and a dihybrid segregation involving the three groups, deep purple, purple and green shoots, was obtained. Twenty-three selections were carried forward from this F₂ generation and an F₃ population raised. In the F₃ generation, the dihybrid segregation of the F₂ together with other simple monogenic segregations were obtained. These are given in Table IV :—

TABLE IV
Segregating for PC_1 and PJ
(Shoots deep purple, purple and green)

Generation	Family No.	Character of selection	Behaviour of progeny		
			Shoot colour		
		Shoot colour	Deep purple	Purple	Green
F_2	M S 1428 family				
	A S 3949	Deep purple . . . Expected 9 : 3 : 4 ratio $\chi^2 = 1.6084$	53 47.25	13 15.75 $P > 0.30$	18 21
F_3	A S 4812	Deep purple . . .	229	59	91
	" 4813	" " " "	236	81	108
	" 4814	" " " "	215	65	76
	" 4815	" " " "	291	107	119
	" 4820	" " " "	225	67	98
	" 4832	" " " "	63	17	27
		Total . . .	1259	396	519
		Expected 9 : 3 : 4 ratio $\chi^2 = 2.5001$	1229.9	407.6 $P > 0.20$	543.5
	A S 4816	Deep purple . . .	364	121	..
	" 4817	" " " "	138	31	..
	" 4818	" " " "	389	153	..
	" 4819	" " " "	329	81	..
		Total . . .	1220	386	..
		Expected 3 : 1 ratio $\chi^2 = 0.7978$	1204.5	401.5 $P > 0.30$..
	A S 4834	Deep purple . . .	383	..	125
	" 4835	" " " "	136	..	47
		Total . . .	519	..	172
		Expected 3 : 1 ratio $\chi^2 = 0.00108$	518.25	.. $P > 0.95$	172.75
	A S 4822	Deep purple . . .	Pure
	" 4823	Purple	243	67
	" 4824	" " " "	..	346	96
	" 4825	" " " "	..	212	68
	" 4826	" " " "	..	261	91
	" 4827	" " " "	..	Pure	..
		Total	1062	322
		Expected 3 : 1 ratio $\chi^2 = 2.2196$..	1038 $P > 0.10$	346
	A S 4828	Green . . .	"	"	Pure
	" 4829	" " " "	"	"	"
	" 4830	" " " "	"	"	"
	" 4831	" " " "	"	"	"
	" 4836	" " " "	"	"	"

GENETIC ANALYSIS OF THE ROOT PIGMENTATIONS

The inheritance of the root pigmentations has been studied in 21 crosses. Segregations up to the third generation were pursued in 11 of these.

The factor **P** [Rangaswami Ayyangar, 1938] is present in the purple group and absent in the brown. Purple is a simple dominant to brown. The purple group is divisible into two sub-groups, one with the factor **Q** [Rangaswami Ayyangar, 1938] which makes the roots reddish purple and the other without it which results in the roots appearing blackish purple. The former is dominant to the latter. These two factors **P** and **Q** result in the following genic constitutions of the three groups.

1. Reddish purple **PPQQ**
 2. Blackish purple **PPqq**
 3. Brown **ppQQ**
- or **ppqq**

The interplay of these two factors results in the usual monohybrid and dihybrid ratios. Segregations for these three root pigmentations are presented in the following tables. Tables V-VII give simple monogenic segregations for one or the other of the two factors. Table VIII gives the dihybrid segregation.

TABLE V
Pure for q and segregating for P
(Roots blackish purple to-Brown)

Generation	Family No.	Character of selection	Behaviour of progeny	
		Root colour	Root colour	
			Blackish purple	Brown
Parents	A S 4068	Blackish purple		
	" 2380	Brown		
F ₁	" CCLXXVIII	Blackish purple		
F ₂	" 6411	"	355	120
	" 6412	"	210	75
	Total		565	195
	Expected 3 : 1 ratio		570	190
			$\chi^2 = 0.1754$	$P > 0.50$
F ₃	A S 6451	Blackish purple	320	112
	" 6452	"	272	87
	" 6454	"	286	93
	" 6455	"	315	107
	" 6456	"	292	102
	" 6457	"	198	63
	" 6459	"	295	98
	" 6460	"	356	121
	" 6453	"	Pure	..
	" 6458	"	"	..
	Total		2334	783
	Expected 3 : 1 ratio		2337.75	779.25
			$\chi^2 = 0.0240$	$P > 0.80$

The same experience was met with in 18 other families. Three of these gave a total of 410 plants with blackish purple and 145 plants with brown root in the F_2 generation and the other 15 gave a total of 1305 blackish purple and 384 brown rooted plants in the F_3 generation.

TABLE VI
Pure for Q and segregating for P
(Roots reddish purple to brown)

Generation	Family No.	Character of selection		Behaviour of progeny	
		Root colour		Root colour	
				Reddish purple	Brown
Parents	A S 3834	Reddish purple			
	" 2487	Brown			
F_1	" CCLXXIII	Reddish purple	.		
F_2	" 5272	"	.	287	99
	" 5273	"	.	156	56
	" 5274	"	.	325	115
	" 5275	"	.	205	69
		Total	.	973	339
		Expected 3 : 1 ratio		984	328
				$\chi^2 = 0.4918$	$P > 0.30$
F_3	A S 6067	Reddish purple	.	200	68
	" 6071	"	.	270	88
	" 6072	"	.	175	62
	" 6068	"	.	Pure	..
	" 6069	"	.	"	..
		Total	.	645	218
		Expected 3 : 1 ratio		647.25	215.75
				$\chi^2 = 0.0312$	$P > 0.80$

The same experience was met with in 11 other families. Six families gave 2343 reddish purple and 922 brown in the F_2 and the other five families gave 1316 reddish purple and 483 brown in the F_3 generation.

TABLE VII

Pure for P and segregating for Q

(Roots reddish purple to blackish purple)

Generation	Family No.	Character of selection	Behaviour of progeny	
		Root colour	Root colour	
			Reddish purple	Blackish purple
Parents	A S 4249	Reddish purple		
	" 29	Blackish purple		
F ₁	" CCLXVI	Reddish purple		
	" 5429	435	138
F ₂	" 5430	392	126
		Total	827	264
		Expected 3 : 1 ratio	818.25	272.75
		$\chi^2 = 0.374$	$P > 0.50$	
Parents	A S 6403	Reddish purple	151	45
	" 6306	"	65	25
F ₁	" 6307	"	64	23
	" 6305	"	Pure	"
F ₂	" 6308	Blackish purple	..	Pure
		Total	280	93
		Expected 3 : 1 ratio	279.75	93.25
		$\chi^2 = 0.0008$	$P > 0.95$	

The same experience was met with in 73 other families. Of these 21 families gave a total of 5537 reddish purple and 1861 blackish purple in the F₁ and the rest 47 families gave a total of 12,489 reddish purple and 4,123 blackish purple in the F₂ generation.

The dihybrid segregation involving the factors **P** and **Q** was pursued in crosses AS CCLXXXII each parent contributing one dominant gene. The parents involved are AS 4564, a blackish purple root selection and consequently of the constitution **PPqq**, and AS 2487, a brown root selection of constitution **ppQQ**. In the F₁ generation both the dominant genes are brought together resulting in the reddish purple roots. In the F₂ generation dihybrid ratio was obtained. From the F₂ generation a number of selections were carried forward to a third generation and as expected both the dihybrid and monohybrid segregations were obtained. This experience is given in Table VIII.

TABLE VIII

Segregating for P and Q

(Roots reddish purple, blackish purple and brown)

Generation	Family No.	Character of selection	Behaviour of progeny		
		Root colour	Root colour		
			Reddish purple	Blackish purple	Brown
Parents	A S 4564 . " 2487	Blackish purple . Brown			
F ₁	„ CCLXXXII	Reddish purple .			
F ₂	„ 6153 .	„ .	268	84	101
	„ 6154 .	„ .	302	98	126
	„ 6155 .	„ .	283	97	125
		Total . Expected 9: 3 : 4 ratio	853 834.75	279 270.25	352 371
			$\chi^2=1.3739$ $P>0.50$		
F ₃	A S 6462 .	Reddish purple .	165	45	63
	„ 6463 .	„ .	71	26	34
	„ 6464 .	„ .	63	22	31
		Total . Expected 9: 3 : 4 ratio	299 292.5	93 97.5	128 130
			$\chi^2=0.3828$ $P>0.80$		
	A S 6465 .	Reddish purple . Expected 3 : 1 ratio	120 117	36 39
			$\chi^2=0.3077$ $P>0.50$		
	A S 6461 .	Reddish purple . Expected 3 : 1 ratio	153 151.5	49 50.5
			$\chi^2=0.0594$ $P>0.80$		
	A S 6466 .	Brown	Pure
	„ 6467 .	„	„

The same experience was met with in 28 other families. Sixteen of these gave a total of plants with 3778 reddish purple, 1215 blackish purple and 166 brown roots in the F₂ generation and the rest 12 families gave a total of 306 reddish purple, 980 blackish purple and 1354 brown roots in the F₂ generation.

INTER-RELATIONSHIP OF SHOOT AND ROOT PIGMENTATION GENES

The inheritance of the factor pairs **PCPC** and **PJPJ** determining the shoot pigmentations, and of the factor pairs **PP** and **QQ** determining the root pigmentations have been discussed in the previous pages. The inter-relationship of these four factor pairs will be dealt with in this part. The cross collations and linkages discussed in this part are from artificial crosses designed for the purpose. The linkage relationships are studied both in the coupling and in the repulsion phases and the segregations are pursued up to the third generation. The results are discussed below.

The factor pair **PCPC** is independent of **QQ** and is linked with **P** with a cross-over value of 18.0 per cent. The factor pair **PJPJ** is independent of **Q** and **PP**. Hence it follows that these four pairs of genes are distributed in three pairs of chromosomes, **PP** and **PCPC** being located in one and the same chromosome. Data relating to the above relationship is presented in Tables IX—XIII.

TABLE IX

Crosses with the factor pairs QQ and PC PC

Generation	Family No.	Behaviour of the progeny				
		Shoot	Purple		Green	
		Root	Reddish purple	Blackish purple	Reddish purple	Blackish purple
Parents	A S 367 .		♀	—	—	—
	" 3464 .		..	—	—	♂
F ₁	" CCVIII		F ₁			
F ₂	" 4391 .		385	128	131	39
	" 4393 .		365	124	122	38
	" 4394 .		392	127	142	42
	" 4396 .		420	134	124	30
	Total .		1562	513	529	149
Expected 9 : 3 : 3 : 1 ratio			1548.56	516.19	516.19	172.06
			$\chi^2 = 3.5447$		$P > 0.30$	
F ₃	A S 5341 .		165	52	58	21
	" 5346 .		301	95	101	35
	" 5349 .		256	82	87	30
	" 5350 .		360	111	122	41
	" 5352 .		156	51	56	20
	" 5353 .		115	41	39	12
	" 5363 .		260	82	93	25
	" 5364 .		160	56	52	18
	" 5365 .		143	42	47	18
	" 5368 .		138	43	41	17
	" 5370 .		24	7	7	3
	Total .		2078	662	703	240
Expected 9 : 3 : 3 : 1 ratio			2071.69	690.56	690.56	230.19
			$\chi^2 = 1.8425$		$P > 0.50$	

The above experience shows that **PC** is independent of **Q**. Similar dihybrid segregations are obtained in six other families and the total figures are :

Shoot	Purple		Green	
Root	Reddish purple	Blackish purple	Reddish purple	Blackish purple
Expected	1244	392	385	120
9 : 3 : 3 : 1	1234.31	411.44	411.44	133.81
		$\chi^2 = 4.1187$	$P > 0.20$	

TABLE X

*Crosses with the factor pairs **PJ PJ** and **QQ***

Generation	Family No.	Behaviour of the progeny				
		Shoot	Deep purple		Purple	
		Root	Reddish purple	Blackish purple	Reddish purple	Blackish purple
Parents	A S 3447	—	—	♀	..	—
	„ 60	—	—	—	♂	—
F ₁	„ CCXV	F ₁				
F ₂	„ 4756	356	118	121	46	
	„ 4757	257	91	95	32	
	Total	613	209	216	78	
Expected 9 : 3 : 1 ratio		627.8	209.2	209.2	6.98	
		$\chi^2 = 1.5402$	$P = 0.50$			
F ₃	A S 4850	170	50	59	18	
	„ 4961	71	23	20	7	
	„ 4962	236	98	94	34	
	Total	477	171	173	59	
Expected 9 : 3 : 3 : 1 ratio		495	165	165	55	
		$\chi^2 = 1.5514$	$P > 0.50$			

From Table X it is evident that **PJ** is independent of **Q**. Similar dihybrid segregations were obtained in five other families and the total of these comes to :

Shoot	Deep purple		Purple	
Root	Reddish purple	Blackish purple	Reddish purple	Blackish purple
Expected	460	136	134	45
9 : 3 : 3 : 1	435.94	145.31	145.31	48.44
		$\chi^2 = 3.0482$	$P > 0.30$	

TABLE XI

*Segregations with the factor pairs **PJPJ** and **PP***

Family number	Behaviour of the progeny				
	Shoot	Deep purple		Purple	
	Root	Reddish purple	Brown	Reddish purple	Brown
Natural crosses from Family					
A S 1933—					
.. 4774 . . .		375	120	128	41
.. 4775 . . .		410	135	138	48
.. 4776 . . .		270	85	88	25
Total .		1055	340	354	114
Expected 9 : 3 : 3 : 1 ratio		1047·94	349·31	349·31	116·44
		$\chi^2 = 0.4096$		$P > 0.90$	

From Table XI it is seen that **PJPJ** and **PP** are independent.

TABLE XII

*Crosses with the factor pairs **PCPC** and **PP***

Generation	Family No.	Behaviour of progeny				
		Shoot	Purple		Green	
		Root	Purple	Brown	Purple	Brown
Parents	A S 2487		—	♀	—	—
	.. 1745		—	♂	—	—
F ₁	.. CCLXXV		F ₁	—	—	—
F ₂	.. 5276 .		213	98	90	3
	.. 5277 .		286	125	122	4
	.. 5278 .		550	240	256	8
	Total .		1049	463	468	15
Calculated 9 : 3 : 3 : 1 ratio			1122·2	374·1	374·1	124·6
			$\chi^2 = 141.8088$		$P < 0.01$	

The high value of χ^2 shows that the distribution of the four groups not conform to the normal dihybrid ratio. The **P** value is much below Such a distribution of the four groups as that obtained above can be explained only on the assumption of a linkage between the factor **PC** for purple colour and the factor **P** for the purple root colour. On this assumption the recombination percentage was worked out and a value of 18.2 ± 1.0 was obtained. On the assumption of linkage with a recombination percentage of 18, the expected distribution of the four groups is as follows :

Shoot	Purple		Green	
	Purple	Brown	Purple	Brown
Numbers obtained	1049	463	468	15
Calculated (18 per cent crossing over)	1013.66	482.59	482.59	16.16
	$\chi^2 = 2.5515$		$P > 0.30$	

The value of $\chi^2 = 2.5515$ with P greater than 0.30 shows that the assumption of linkage with 18 per cent crossing over satisfactorily explains this deviation from the usual simple dihybrid ratio.

From the family A S 5276, two more selections were carried forward and a third generation raised. Both these selections segregated repeating the experience of the F_2 generation. On the assumption of the same linkage as in the F_2 with a recombination value of 18 per cent the distribution of the four groups is as follows :—

Shoot	Purple		Green	
	Purple	Brown	Purple	Brown
A S 6080	192	88	82	3
„ 6081	302	150	148	4
Total	494	238	230	7
Calculated (18 per cent crossing over)	492.35	234.40	234.40	7.15
	$\chi^2 = 0.2353$		$P > 0.95$	

The above linkage between the factor pairs **PCPC** and **PP** is in the repulsion phase. The same experience was obtained in the coupling phase also, and the results are given in Table XIII.

TABLE XIII
Crosses with the factor pairs **PctPct** and **PP**

Behaviour of progeny						
Generation	Family No.	Shoot	Purple		Green	
		Root	Purple	Brown	Purple	Brown
F ₁	A S 60 .	—	♀	—	—	—
	„ 4174 .	—	—	—	—	♂
	„ CCLVIII		F ₁	—	—	—
	„ 5420 .		442	69	65	120
	„ 5421 .		495	70	65	130
	Total .		937	139	130	250
Calculated (18 per cent crossing over) .			972·75	119·25	119·25	244·75
			$\chi^2 = 5·6662$		$P > 0·10$	

From family A S 5420 two more selections were carried forward and a 2nd generation raised. The results are given below :—

Shoot		Purple		Green	
Root		Purple	Brown	Purple	Brown
5296	· · / · ·	168	34	35	55
5297	· · · · ·	275	35	30	61
Total		443	69	65	116
Calculated (18 per cent crossing over)		462·99	56·76	56·76	116·49
		$\chi^2 = 4·7006$		$P > 0·10$	

From the above experience it is evident that the factor **PC** for the purple colour of the shoot and the factor **P** for the purple colour of the root are linked with a crossing-over percentage of 18.

PIGMENTATION OF THE MESOCOTYL

A feature in which grass seedlings differ from those of other mono-cotyledons in general is the frequent presence of a segment inserted between the cotyledon sucker and the sheath which encloses the plumular bud. This segment is called the mesocotyl [Arber, 1934].

In sorghum, the mesocotyl grows to remarkable lengths and is the organ responsible for the emergence of the young shoot from the soil. When the seeds are sown near the ground level, the mesocotyl is short but when they are sown deep, the mesocotyl elongates and thus enables the shoot to emerge out of the soil [Rangaswami, Vidyangar and Nambiar, 1934].

Under normal conditions the mesocotyl is always below the soil and it is found that in such cases it always develops the colour of the roots. If the roots are purple, the mesocotyl is purple. If the roots are brown, the mesocotyl is likewise brown. It is noticed that long before the manifestation of the pigment in the roots, the mesocotyl develops the colour and is always an easy guide for determining the root pigmentation. The next step was taken to grow the mesocotyl above the soil in presence of light like the shoot. For this purpose, seeds were sown in trays filled with soil and grown in darkness for two to three days. The seedlings in darkness became etiolated and the mesocotyl region grew out of the soil to a very long length. When examined, no trace of the pigment was seen either in the young shoot or in the mesocotyl. The trays were then taken out from darkness and exposed to light. The shoots slowly developed their corresponding pigmentation. It was very interesting to observe that the mesocotyl region also along with the shoot began to develop the shoot pigmentation. If the shoots were deep purple, the mesocotyls developed deep purple colour. If the shoots were purple, the mesocotyls were purple. In green shoots the mesocotyls remained colourless.

Thus it is seen that the mesocotyl which under normal conditions is always below the soil and possesses the root colour when made to grow above the soil in presence of light develops the shoot colour. This differential response of this organ to the development of the pigment under different environmental conditions, behaving sometimes as shoot and sometimes as root, is very interesting and is probably brought about as a result of the change in function of this organ under the different environmental conditions.

SUMMARY

The sorghum plant manifests several types of pigmentation in the seedling stage. Both the shoot and the roots develop different types of pigmentation. A classification of the seedlings is made based on both these shoot and root pigmentations.

There are three types of shoot pigmentations, deep purple, purple and green. In the deep purple type, the coleoptile and the young shoot close to its emergence from the soil are coloured deep purple, and in the seedling, the auricular leaf-junctions and the outer margins of the leaf-sheaths are coloured purple. In the purple type, the coleoptile and the young shoot are coloured purple, while the auricular junctions and the leaf-sheath margins are not purple pigmented. In the green type, the coleoptile, the young shoot, the auricular junctions and the leaf-sheath margins are green without any purple in them.

There are three types of root pigmentations, reddish purple, blackish purple and brown. These colours manifest when the seedlings are about 10 or more days old. The reddish purple roots are brick-red with a tinge of purple colour in them. When dried they lose the purple tint and become brown.

k-red. The blackish purple roots are likewise black with a slight tint of purple. On drying the purple tinge disappears and the roots ultimately become black. In the brown type the roots are yellowish brown when fresh and on drying become brown to straw in colour.

Based on these shoot and root pigmentations, sorghum seedlings are classified into nine pigmentation groups:—

These seedling pigmentations have a definite relationship with some of the adult plant pigmentations. When the shoots are deep purple, the leaf-blades are likewise coloured deep purple and the leaf-junctions, the leaf-sheath margins and the nodal bands are coloured purple. When the shoots are purple, the leaf-axils alone are coloured purple and the other regions are green. When the shoots are green, all these regions including the leaf-axils are green with any trace of purple in them. When the roots are coloured purple, the leaf-blades and the glumes of the plant are also purple. When the roots are brown, the leaf-sheaths and glumes are likewise brown. The reddish purple roots are associated with reddish purple leaf-sheaths and glumes, and the blackish purple roots with blackish purple leaf-sheaths and glumes. This relation between seedling and the adult pigmentation is absolute and probably due to the action of the same genes. The mesocotyl develops either the root or the shoot pigmentation according as it is under the soil like the roots or grows above the ground (normally rare) like the shoot.

In inheritance, a factor **PC** is responsible for the purple pigmentation of the young shoot. In its absence (**pc**) the shoot is green. A factor **PJ** acting in the presence of **PC** makes the shoot deep purple and colours the leaf-junctions and leaf-sheath margins purple. **PJ** has no visible effect in the absence of **PC**. The factor **P** is present in the purple-coloured roots and absent in the brown. The factor **Q** makes the roots reddish purple; **q** results in blackish purple roots. The factor **PC** is independent of **Q** and is linked with **P** with a cross-over value of 18 per cent. The factor **P** is independent of **PJ** which again is independent of **Q**.

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EXPLANATION OF PLATE IX

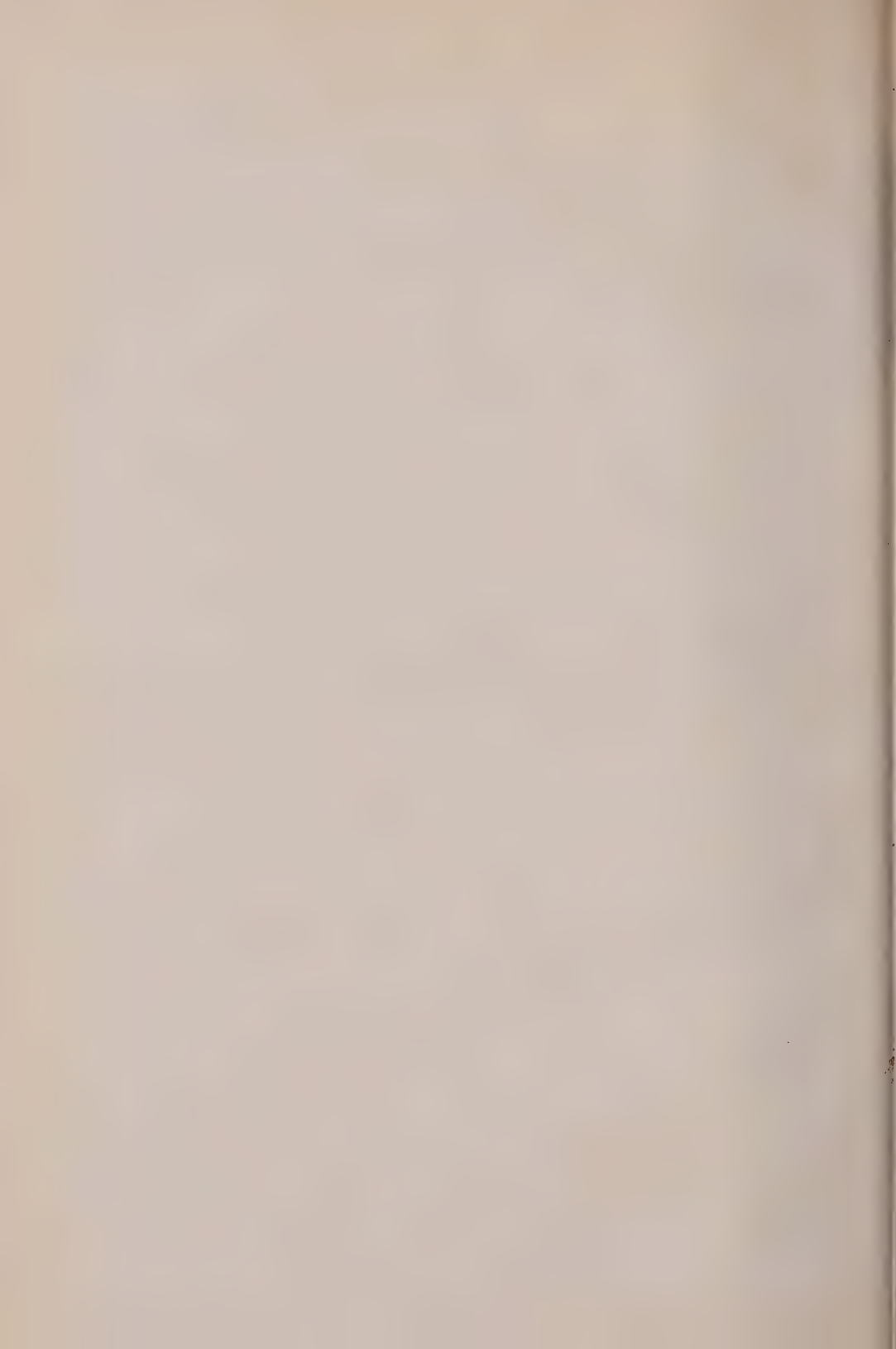
The plate is divided into nine sections, numbered 1 to 9. In these nine sections are shown the nine pigmented types of sorghum seedlings and the related pigmented parts of the adult.

Section No.	Seedlings					
	Shoot					Roots
1	Deep purple	Reddish purple
2	Purple	" "
3	Green	" "
4	Deep purple	Blackish purple
5	Purple	" "
6	Green	" "
7	Deep purple	Brown
8	Purple	" "
9	Green	" "

In each section, A represents a seedling five days old ; B leaf-axil ; C leaf-junctions ; D roots of a seedling 20 days old ; E internode with the leaf-sheath at maturity showing the nodal bands ; and F mature glumes enclosing a well-developed grain.



[For explanation see opposite page



In the deep purple shoot type (Nos. 1, 4 and 7), the coleoptile and the plumule of the shoot A, the leaf-axil B, the leaf-junctions C and the nodal bands in E, are pigmented purple. In the purple shoot type (Nos. 2, 5 and 8), the coleoptile is light purple and the plumular leaves are merely tinged purple; the leaf-axil is light purple, while the leaf-junctions and the nodal bands are green. In the green shoot type (Nos. 3, 6 and 9) the coleoptile, the plumular leaves, the leaf-axil, the leaf-junctions and nodal bands are without any trace of purple in any one of these parts.

In the reddish purple root type (Nos. 1, 2 and 3), the seedling roots D, the mature roots E and the glumes F, are pigmented reddish purple. In the blackish purple root type (Nos. 4, 5 and 6), all these parts are pigmented blackish purple, and in the brown root type (Nos. 7, 8 and 9), these are brown.

EFFECT OF COTTON SEED DISINFECTION ON YIELD

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DAMPING-OFF and seedling-blight diseases of cotton are caused in the provinces by *Pythium* sp., *Macrophomina phaseoli* Ashby, *Rhizoctonia solani* Kuhn., *Rhizoctonia* sp., *Sclerotium Rolfsii* Sacc. and *Colletotrichum indicum* Dast. These fungi often cause poor germination and considerable loss of seedlings. Trials were carried out for the control of these diseases by treating the seed. During the trials it was found that the incidence of these diseases is not annual and not always of the same intensity. The predisposing factors are excessive moisture caused by heavy rainfall and water logging during the critical period, the first three or four weeks after germination, when the seedlings are most susceptible to infection by these fungi. In the case of the anthracnose disease (*Colletotrichum indicum*) of seedlings the use of infected seed. For these reasons the value of seed treatment for the control of these diseases is not well established except in the case of seed-borne anthracnose disease [Dastur, 1934]. However, there is evidence that the mortality of seedlings raised from disinfected seeds is less than that of seedlings from untreated seeds in years when there is an epidemic of these diseases. But these trials have shown the value of seed disinfection in increasing the yield.

In Nagpur, these trials have been carried out since 1936 and for the two years these treatments have been tried on some of the district farms as well. The seed treatments tried at Nagpur were dusting with Agron G, Hortosan B, Abavit B, Ceresan, finely powdered copper carbonate sulphur, and delinting with sulphuric acid. The four proprietary fungicides were used at the rate of one ounce per 28 lb. of seed; the two chemicals were used at the rate of two ounces per 28 lb. of seed; for delinting the seed with sulphuric acid one part by volume of the acid was used for 20 parts volume of the seed.

It is a general practice amongst cultivators of the province to wet cotton seed with a cowdung solution so that, on drying, the fuzz is set and the seed passes readily through the drill. The fungicide in the powder was well mixed with the cowdung solution before it was applied to the seed.

Delinting with sulphuric acid was carried out as described in a previous publication [Dastur, 1934].

In 1936 each treatment was tried on 1/40 acre plot and replicated three times. Each block consisted of these seven treatments and one control. The three blocks were placed contiguously. The following year the trials were repeated; each treatment and the control were tried on 1/40 acre plots and replicated three times. In 1938, these trials were again carried out with the same number of replications but each plot was 1/20 acre.

It was observed that the delinted seeds germinated earlier by a day or two than the other seeds. The plants in the control plots were at first smaller than those in plots sown with treated seed. This difference in growth was noticeable only up to about two months after sowing; but when the flowering stage was reached, there was no visible difference in the size of the plants.

The yield of *kapas* (seed cotton) from the plots sown with treated seed was higher than that from the control plots during these three years.

As the treated seed in the trials had given a higher yield than the untreated seed, it was considered desirable to increase the number of replications and to test only those fungicides which are readily procured in India and which can be easily used by the cultivator. Ceresan is not marketed in India; delinting seed with sulphuric acid cannot be done by the ordinary cultivator; therefore, these two treatments were not tried again. In 1939 and 1940, each plot was 1·20 acre and the replications were six; the treated and untreated seeds were sown in randomized plots. The percentage of increase in yield from the five years' trials are given below:—

Treatment	1936-37	1937-38	1938-39	1939-40	1940-41
Agrosan G . . .	14·4	27·0	38·3	19·0	1·7
Hortosan B . . .	9·1	20·6	44·6	21·1	21·2
Abavit B . . .	8·0	13·9	40·3	17·5	5·9
Ceresan	16·3	25·9	25·9
Copper carbonate . .	25·3	24·9	38·9	8·2	12·7
Sulphur	15·2	29·9	33·7	19·5	12·4
Delinted seed . . .	8·7	9·9	10·4

In 1940, the monsoon was unseasonable and, when it broke, the rains were abnormally heavy and continuous; the result was that all the six blocks could not be sown on the same day; a block and a half were sown on 30 June; further sowing could not be done till 8 July when only 3½ blocks could be sown; the remaining sixth block could not be sown on account of rain. No further sowing was possible till 25 July. The fields got flooded and some of the plots sown on 8 July were under water for many days; either seeds were washed away or seedlings were killed. For these reasons the yield from only a few plots could be considered, those that were sown in the end of June and some of those that were sown on 8 July. Last year's results cannot be therefore compared with those of the previous four years.

In 1939 and 1940, three of these fungicides were tried out on the Akola Farm. Each plot was $\frac{1}{4}$ acre and replicated four times. The plots were randomized. The percentage of increase in yield is given below :—

Treatment	1939-40	1940-41
Agrosan G	11.3	12.5
Copper carbonate	11.8	4.7
Sulphur	10.6	1.9

At the Government Farm, Khandwa, in 1939 seed treated with Agrosan G and copper carbonate were sown on 0.3 acre plots with six replications. The control plot was also replicated six times. In 1940, the size of each of these three plots was $\frac{1}{20}$ acre ; each plot was replicated six times. These trials were carried out in randomized plots. The percentage of increase in yield is given below :—

Treatment	1939-40	1940-41
Agrosan G	26.5	10.4
Copper carbonate	12.4	6.8

On a demonstration plot at Amraoti, Agrosan G and sulphur treatments were tried in 1939 and 1940. The 1940 trials were vitiated on account of the unseasonal and abnormal rainfall, and therefore the results of the 1940 trials cannot be considered. In 1939, seeds treated with Agrosan G and with sulphur were sown on two-acre plots each and the untreated seed was sown on a one-acre plot. The percentage of increase of yield from the plots sown with seed treated with Agrosan G and sulphur was 16.2 and 11.5 respectively.

On the Government Farm at Ellichpur in 1939, a field of 10 acres was divided into four equal plots of $2\frac{1}{2}$ acres each ; one plot was sown with seed treated with Agrosan G, another plot with copper carbonate-treated seed and the remaining two plots were sown with untreated seed and alternated with the plots sown with the treated seed. The yield from the plot sown with seed treated with Agrosan G was 14.7 per cent higher than that from the control plots, but the yield from the plot sown with seed treated with copper carbonate was less by 19.4 per cent,

The following year these two fungicides were tried again on this Farm two fields separated from each other. In one field each plot was 1/3 acre and replicated five times. On the second field these trials were repeated six times in randomized plots of 1/20 acre each. The percentage of increase in yield from these two plots is given below :—

Treatment	Field I	Field II
Grosan G	12.6	7.6
Copper carbonate	7.6	5.3

It is not possible to give a precise explanation for the lower yield obtained from the plot sown with copper carbonate-treated seed in 1939. But since the following year on two fields on the same Farm the yield was higher than from the control plots, it is permissible to assume that the lower yield in 1939 could not be due to the treatment.

In 1940, some of these treatments were tried at Borgaon and Yeotmal farms. Each of the plots sown with treated and untreated seed was 1/20 acre and replicated six times. The percentages of increase in yield are given below :—

Treatment	Borgaon Farm	Yeotmal Farm
Grosan G	2.0	31.2
Copper carbonate	6.8	19.0
Sulphur	9.6	32.5

The results of these experiments, which have been carried out for many years and on various Government Farms, show that chemical treatment of cotton seed increases the yield of *kapas* (seed cotton); the percentage of increase is variable, but is sufficient to justify the use of treated seed; seed treatment is inexpensive and does not need any extra labour or skill or care. The treatment with fungicides not only ensures increase in yield but also is capable of destroying fungus spores, e.g. spores of the anthracnose fungus; this disinfection of the seed would naturally ensure better germination and reduction in loss from disease, especially when conditions are unfavourable for quick germination.

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STEM-BROWN DISEASE OF APPLE IN KUMAUN

BY

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(With Plates X-XIV)

A SERIOUS disease of apple stem caused by *Botryosphaeria ribis* Gross and Dugg. was recorded for the first time in India in August 1934 in the Government Orchard, Chaubattia (Kumaun), and a short note regarding the disease was given in the Annual Report of the Hill Fruit Research Scheme, Chaubattia for the year 1934-35. The specimen was sent to Mr. Ashby, the Director of the Imperial Mycological Institute, Kew, who identified it as *Botryosphaeria ribis* Gross. and Dugg. and he enquired whether the fungus in culture formed a red colour on starch or not, which the parasitic strains of *B. ribis* were known to produce. Since it produced this colour on starch in cultures, the fungus proved to be identical with *B. ribis*. This fungus has had a great variety of names applied to it in its different stages. The ascogoneous stage is best known in Europe under the name of *Botryosphaeria Berengerians* de Not. In America it has been frequently called *Botryosphaeria fuliginosa* (M+N) E & E. The occurrence in the Hudson Valley of a destructive blight or wilt of currants was first described by Fairchild [1891] and ascribed to a 'sterile' fungus. The importance of *Botryosphaeria ribis* as a pathogene of currant and gooseberry was demonstrated by Grossenbacher and Duggar [1911]. They described the perfect stage as *B. ribis*. The imperfect stage common on many hosts is a *Dothiorella*. Putterill [1919] described a canker of apple trees in South Africa, which was caused by a fungus closely resembling *Botryosphaeria ribis*. He called it *Botryosphaeria mali*, the difference between it and *B. ribis* being in the width of asci and in the size of the stroma. Recently Stevens and Jenkins [1924] have shown that the fungus occurs on horse chestnuts and is parasitic on roses causing canker on the stem and sometimes killing the whole canes. An exhaustive history of the fungus and its host range is given by Smith [1934] who found the fungus to have a wide host range, including at least 34 genera and 20 families of plants, one of which is the apple. Birmingham [1924] recorded *Dothiorella mali* as causing a stem disease of apples in Australia. *Botryosphaeria ribis*, thought to be the perfect stage of *D. mali* was found to cause rotting of apples by Fenner [1925]. Hopkins and Bacon [1938] recorded this disease on apple stem in Southern Rhodesia.

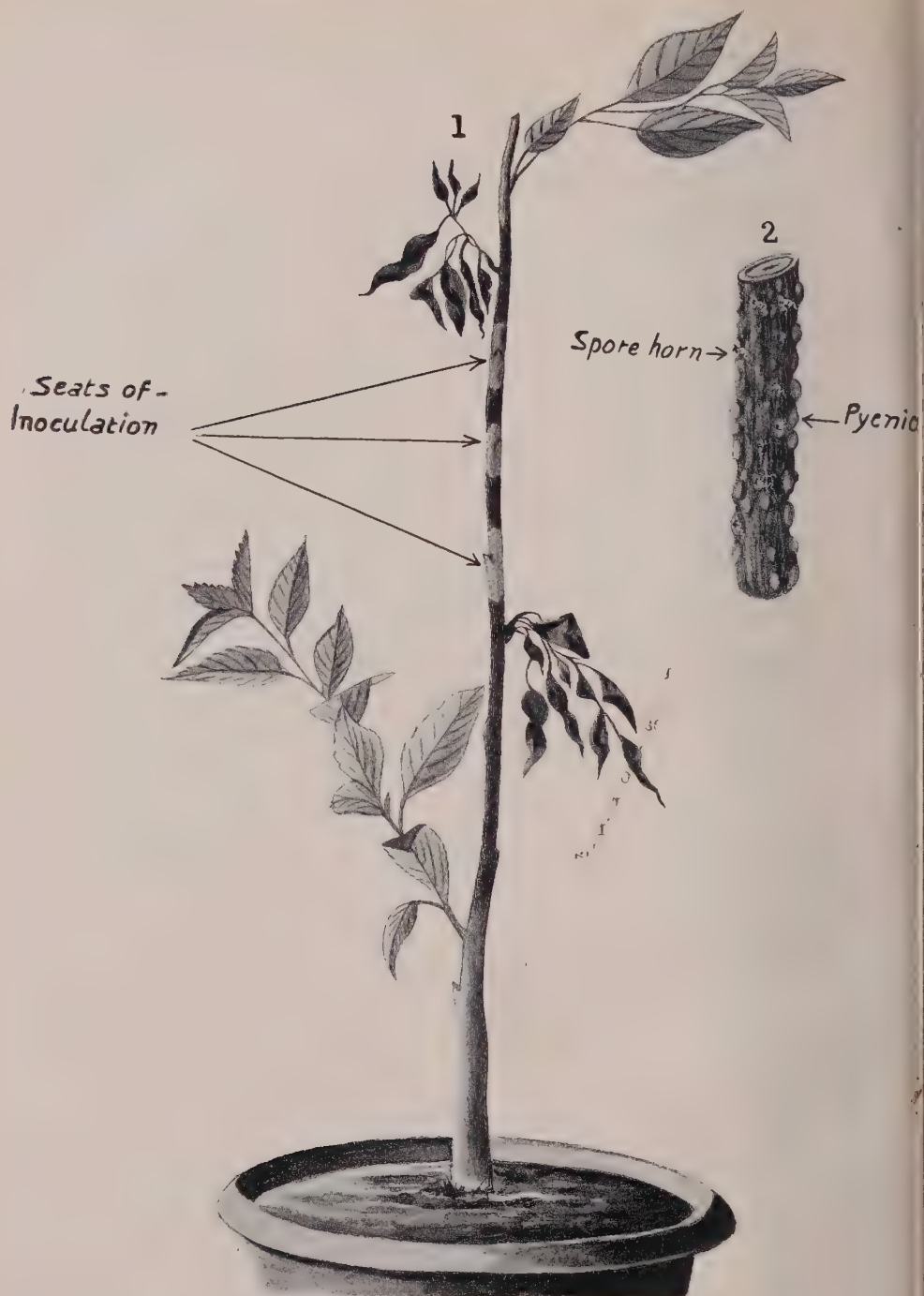
SYMPTOMS OF THE DISEASE

The disease starts from the pruned surface of twigs and stems and proceeds downwards causing a type of die-back (Plate X, fig. 1). The upper limbs of the apple trees are more liable to the attack. It causes a loosening

STEM-BROWN CAUSED BY *Botryosphaeria ribis*



1. Stem-brown disease on a young shoot ($\times \frac{1}{3}$)
2. Stem-brown disease on old shoot showing pycnidia ($\times 1\frac{1}{3}$)
3. Pycnidia enlarged ($\times 2\frac{2}{3}$)
4. Stem-brown disease on an old shoot showing perithecia ($\times 1\frac{1}{3}$)
5. Stem-brown disease on perithecia enlarged ($\times 2\frac{2}{3}$)

PYCNIDIAL STAGE OF *Botryosphaeria ribis*

1. One-year-old plant of *Esopus Spitzenberg* artificially inoculated at three places ($\times \frac{1}{4}$)
2. An enlarged portion of the infected stem showing pycnidia 'B' type with spore horns (\times)

the bark which becomes papery and brown and rolls outwards (Plate X, fig. 1). On removing the bark, the wood is found to be stained dark brown colour and fissured both horizontally and longitudinally (Plate X, fig. 2). A combined attack of the fungus and *Coniothecium chomatosporum* Corda, which caused another similar disease, is often found to be present in the cankered wood. The disease is noticed usually by the fourth week of April and is in its most virulent form by the middle of May. In old herbarium specimens pycnidia and perithecia are met with. Perithecia are rarely found in nature. The mycelium of the fungus is found right up to the wood and is dark brown in colour, closely septate tending to form chlamydospores. The fruiting bodies, especially pycnidia of simple type, are formed just outside the cork-forming layer which is depressed by their growth. The primary excrescence is raised to form marked protuberances. Compound pycnidia form small globular sclerotial bodies between the phellogen and outer cortical parenchyma of shoots and stems.

MORPHOLOGY OF THE FUNGUS

PYCNIDIA

Two types of pycnidia are met with in the old diseased specimens of the apple stem. For convenience these two types will be referred to in this paper as pycnidia-A and pycnidia-B.

Pycnidia-A

They are very minute structures dotted all over the stem. Their conceptacles are sclerotoid, immersed, crowded with distinct ostioles in the mature stage and measure 0.25-0.5 mm. There is a small stroma which is sometimes confluent. The central core of the conceptacle remains as solid pseudoparenchyma of thick-walled, soft cells, and around this in the hyaline part of the stroma irregular cavities are formed (Plate XII, figs. 4 and 5). These cavities become irregularly lined with pycnosporo-mother-cells which form within them bacillar, allantoid spores measuring $9-12 \times \pm 1\mu$ which are hyaline and apparently continuous. The spores seem to be set free by the dissolution of the spore-mother-cells and are thus endogenous.

The pycnospores are regarded as spermatia, and all attempts to germinate them have been unsuccessful. There are some points of resemblance of these to the filiform bodies figured by Klebhan [1933] for *Sclerophoma strobili*. This pycnidial stage of the fungus may well be termed the sclerophoma stage. These pycnidia-A are formed in the vicinity of stromata destined to give rise to perithecia. Spear [1910] also described a similar type of small, hyaline, cylindrical pycnospores measuring $2-3\mu \times 1\mu$ which he found on the host alive and not in culture.

Similar structures were also described by Grossenbacher and Duggar [1911], but they considered that these did not belong to the life-cycle of *Dothiorella ribis*. The spores failed to germinate. Tulasne and Tulasne [1863] described and figured a similar stage for their *Dothiorella melanops*, now usually called *B. melanops* (Tul.).

Pycnidia-B

In nature pycnidia-B are found in greater abundance than pycnidia-A. They are minute, ostiolate bodies (Plate X, fig. 3) and at times give out spores. The spores are light pink in colour. They are either simple or compound stylospore form (*Dothiorella*) and are borne in the same or similar stroma as the perithecia.

Pycnidia-B are either single or in groups of two to six, globose, black, erumpent, ostiolate having a thick wall (Plate XII, fig. 1). They measure $126-394 \times 114-8-210 \mu$; the average size being $220-7 \times 166-2 \mu$. The conidiophores are small, unbranched, hyaline, bearing pycnosporangia at the tip. The pycnosporangia are fusoid to oblong, elliptical, hyaline to subhyaline, unicellular and measure $9-1-25-6 \times 5-6-7-7 \mu$; average $17-5 \times 6-8 \mu$ (Plate XII, figs. 3a-3c). These pycnosporangia germinate readily in tap water within 8-12 hours and send out germ tubes from either end (Plate XII, figs. 6a-6f).

PERITHECIA

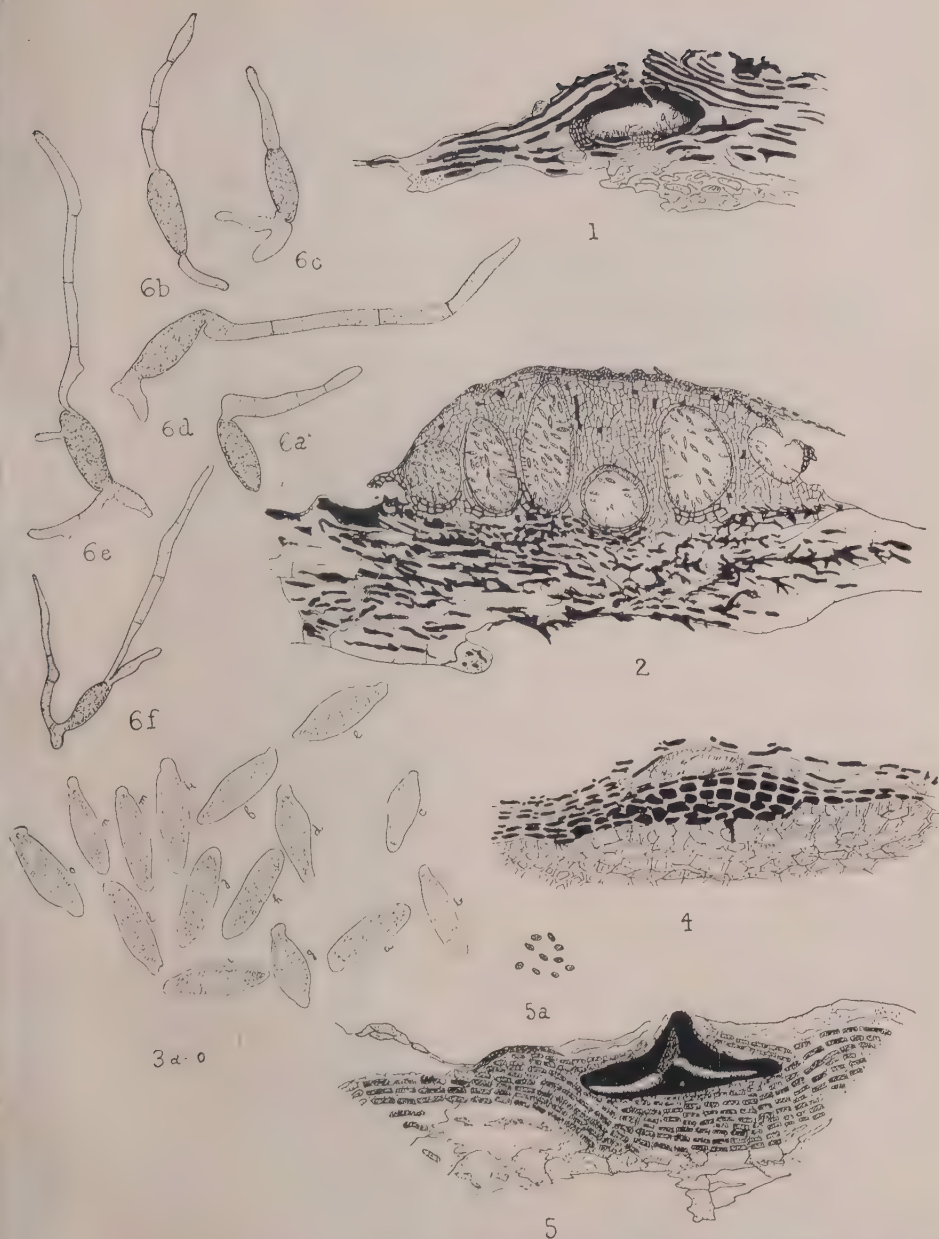
The perithecia are not so commonly met with in nature as the pycnidia. They are round, top-shaped bodies with papillate ostioles (Plate X, figs. 4 and 5). They either occur singly or in groups of two to eight (Plate XI, figs. 1 and 2). The stroma are black, more or less pulvinate, and measure from 0.5 to 5 mm. in diameter. The perithecia are sometimes interspersed among pycnidia. The perithecia measure $140-0-252 \times 127-4-280 \mu$; average $190-7 \times 192-0 \mu$. The asci are clavate, eight spored, hyaline and measure $36-4-112-0 \times 14-0-18-9 \mu$; average $77-5 \times 15-4 \mu$ (Plate XI, figs. 3a, 4 and 5a-5d). The ascospores are biserial in arrangement, unicellular, hyaline, fusoid, elliptical to ovoid (Plate XIII, figs. 6 and 7a-7d) and measure $11-9-28-00 \times 8-4-12-6 \mu$; average $20-2 \times 10-7 \mu$. The paraphyses are present and are filiform. The ascospores germinate in tap water at room temperature in 10-14 hours throwing out germ tubes from either end. (Plate XIII, figs. 8a-8d). Affected pieces of apple twigs bearing pycnidial structures were kept immersed in the snow for one month and, when examined in March, were found to contain mature and immature perithecia. What part the perithecia play in the spread of the disease is difficult to say, but at any rate there is clear indication that old affected twigs of apple bearing pycnidia if buried in the winter snow will undoubtedly be a potential source for the spread of the disease in the following spring.

CULTURAL CHARACTERS

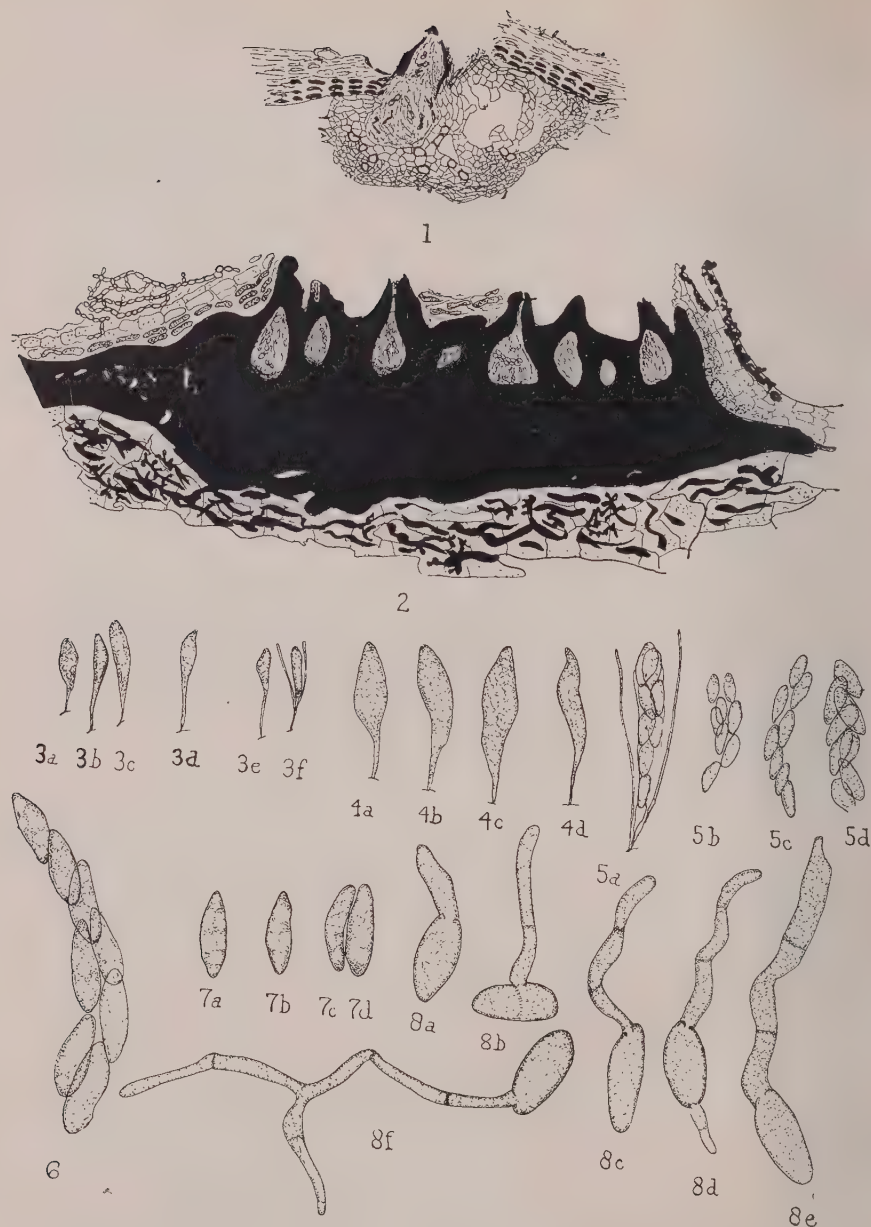
The cultural studies of the three isolates of *Botryosphaeria ribis*, no. XXI (from single pycnosporangium), St-Br (from single ascospore) and 123 (from Holland) (Plate XIV, figs. 1 and 2) were carried out in detail on three media, Czapek's agar, potato-dextrose agar and Brown's starch synthetic agar.

Linear measurements of growth

All the cultures were grown in petri dishes of equal size and the amount of medium in each was 10 c.c. Each treatment was replicated six times and the four measurements of each of the colonies were taken. All the petri dishes were kept at a constant temperature of 30°C. in an incubator.



1. Transverse section of bark of apple stem showing single pycnidia 'B' type (*Dothiorella*) ($\times 107$); 2. Transverse section 8μ thick of bark of apple stems showing pycnidia 'B' of the compound stylosporid form ($\times 107$); 3a-o. Pycnosporangia from pycnidia 'B' type ($\times 1,066$); 4. Transverse section of bark of apple stem showing pycnidia 'A' type (*Sclerophoma* type) ($\times 107$); 5. Transverse section 8μ thick of bark of apple stem showing pycnidia 'A' type (*Sclerophoma* type) ($\times 107$); 5a. Pycnosporangia from pycnidia 'A' type ($\times 1,066$); 6a-f. Pycnosporangia 'B' germinating ($\times 1,066$)



1. Transverse section of bark of apple stem showing perithecia ($\times 107$);
 2. Transverse section of bark of apple stem showing group of perithecia ($\times 107$);
 3a-3f. Immature asci ($\times 480$); 4a-4d. Immature asci ($\times 1,066$); 5a-5d. Mature
 asci with ascospores ($\times 480$); 6. Eight ascospores ($\times 1,066$); 7a-7d. Ascospores
 ($\times 1,066$); 8a-8f. Ascospores germinating ($\times 1,066$)

It was noted that the isolate XXI grows best on Brown's starch synthetic agar, the isolate St-Br, on Czapek's agar and the isolate 123 on potato-dextrose agar. Thus all the three isolates differ from each other as far as the linear rate of growth of the cultures is concerned.

Macroscopic and microscopic characters

Macroscopic and microscopic characters were also noted for the three isolates. All the isolates differ in macroscopic and microscopic characters.

No pycnidia or perithecia were formed by any of these isolates in dark at a temperature of 30°C., but in light, at room temperature, the strain XXI formed pycnidia of 'B' type on potato-dextrose agar only, while the isolate St-Br produced them in all the three media. Photographs of the three isolates growing on potato-dextrose agar are given in Plate XIV, figs. 1a-1c and the paired cultures in petri dishes are shown in Plate XIV, figs. 3a and 3b.

CULTURAL STUDIES OF THE SALTANTS

A saltant appeared in two culture flasks of the local strain No. XXI (Plate XIV, figs. 2a and 2b). They were given the numbers XXIa and XXIaI. They were isolated and grown at 30°C. on Brown's starch synthetic agar, potato-dextrose agar and cornmeal agar for comparison of cultural character with the parent strain No. XXI and also with the strain St-Br. All the cultures were kept in the dark. The principal difference was found to be in the rate of their linear growth which was faster than the parent, but slower than the isolate St-Br. On cornmeal agar and potato-dextrose agar both the saltants produced more broad and distinct zones than the parent isolates. None of them showed zonation on Brown's medium.

INOCULATION EXPERIMENTS

Two isolates of the fungus were taken. One of the strains numbered XXI was isolated from diseased apple twigs bearing pycnidia-B in bacto-agar from the growth of a single germinated pycnospore. The other strain numbered St-Br was isolated from a single ascospore.

The two isolates differ from each other in cultural characters. They both produce the dothiorella stage but no perithecia. A third foreign isolate of *Botryosphaeria ribis* received from Central Bureau Schimmelcultures Baarn (Holland) which was numbered 123 did not produce even pycnidia but remained altogether sterile. All the three strains caused chromogenesis of starch in light.

For establishing the pathogenic characters of the fungus the following experiments were carried out :—

Entry of the fungus through injured surfaces

In April 1937, 12 plants of *Esopus Spitzenberg* grafted on seedling stocks (age of the graft was one year) were selected. In all cases the upper portion of the stem was injured aseptically at three places, and the inoculum was placed over the injury which was then covered over with wet absorbant cotton and finally with cellophane bags (Plate XI, figs. 1 and 2). On the

lower portions of the stem the inocula were placed at three uninjured places and similarly covered. Thus we have the following arrangements :—

1. Plants A, B and C inoculated with culture No. XXI
2. Plants D, E and F inoculated with culture No. St-Br
3. Plants G, H and I inoculated with culture No. 123
4. Plants J, K and L uninoculated controls

All the inoculated and control plants were kept inside a glass cage the air of which was kept constantly humid. After about a month only the injured spots were observed to have taken the infection, while the uninjured ones and the controls remained unchanged.

The inoculated portion at first becomes depressed and becomes light brown in colour. The area of the stem below and above the inoculated portion also turns light brown, its bark becoming thin and papery. In advanced stages the whole of the infected stem turns dark brown and gets studded with innumerable pycnidia which when mature give out light pink spore horns (Plate XI, figs. 1 and 2). They were all B type of pycnidia, belonging to dothiorella stage of *Botryosphaeria ribis*. Pycnidia-A did not appear in any of the infected portions. Perithecia were observed the following year (in December 1938). The progress of infection in all cases was very slow.

CROSS-INOCULATION EXPERIMENTS

Isolate No. XXI was used in the cross-inoculation experiment. The six immature twigs of each of the six pear, peach, apricot and chestnut plants seedling stock (one year old) were cut aseptically with a pair of pruning scissors, inoculated with the mycelium of the fungus and covered over with cellophane bags. In the control, a drop of sterilized water was used instead of the culture. For the sake of comparison the culture of isolate XXI was inoculated on six immature twigs of apples in six plants. All the plants were kept inside a big glass cage and the inside of the cage was kept humid by spraying with water from time to time. The experiment was carried out on April 1938 and the infection was visible after a fortnight in all the inoculated twigs. In each case the fungus was reisolated on oatmeal agar and resembled the isolate culture in all respects. Thus it was proved that the strain No. XXI of *Botryosphaeria ribis* Gross. and Dugg. could easily infect through injured surfaces of twigs of pear, peach, apricot and chestnut. In some cases pycnidia-B appeared on the affected areas of the inoculated twigs.

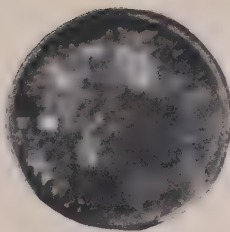
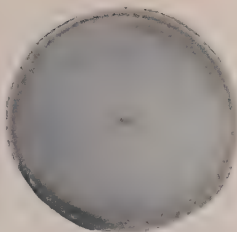
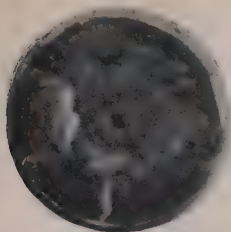
SOURCES OF INFECTION

It was observed that on the already infected shoots pycnidial structures developed by the beginning of April but mature dothiorella type was not found till later. Mature pycnidial structures were only met with in the upper portion of diseased twigs, the lower portion having only immature pycnidia. In the latter part of May, and early June immature perithecia appeared in large numbers among the pycnidia. The dothiorella type of pycnidia, which was more numerous later on in the season continued to ooze out pinkish spore horns till the month of July. Ascospores appeared in June and were copious during the middle and later part of July. Field observations have shown that fresh infections through injured uncallused surfaces of stem and

XXI

123

St-Br.



1a

1b

1c

Cultures No. XXI, St-Br and 123 growing on potato dextrose agar at 30°C. (3 month's old)



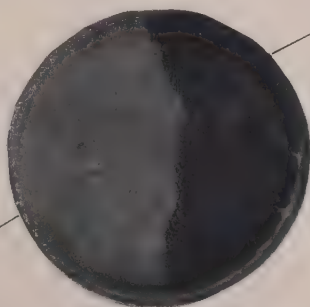
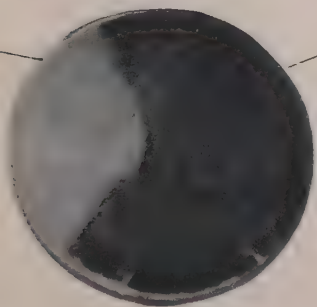
2a

2b

2b. Cultures No. XXI showing white patches of saltant No. XXIa (The culture was growing on ricemeal agar at room temperature)

St-Br.

XXI



3a

123

3b

3b. Paired culture of No. XXI and 123 and St-Br. on Czapek's agar at 30c. (18 days' old)

eggs occur in June and July when the spores were abundantly produced. The diseased areas became prominent in the following season. The spores seemed to be carried by rain water only. What part, if any, insects play in the dissimination of spores was not observed. The pruned diseased twigs and stems left in the orchards also serve as a potent factor in the fresh infections.

METHODS OF CONTROL

As the fungus can only enter through the cut pruned surface of the apple stem, any suitable paint capable of preventing the entry of the fungus will be an effective method of controlling the disease.

Experiments carried out in 1938

In 1938 a control experiment was started to find out a suitable paint for covering the pruned surfaces of apple twigs as a protection against the entry of the fungus. This experiment was done in two sets—one for mature twigs and the other for immature ones. For each set 12 old trees were selected at random spread all over the orchard.

The following six treatments each replicated six times were carried out on each tree :—

- (i) Untreated (uninoculated) control I
- (ii) Untreated (inoculated) control II
- (iii) Treated with red lead-copper carbonate paste followed by inoculation. (The paste was prepared with 100 c.c. of raw linseed oil and 2 oz. each of red lead and copper carbonate.)
- (iv) Treated with red lead-Bordeaux powder paste followed by inoculation. (This paste was prepared with 75 c.c. of raw linseed oil and 2 oz. each of red lead and dried precipitate of 4 : 4 : 50 Bordeaux mixture.)
- (v) Treated with mercuric chloride solution followed by Bordeaux paste and then inoculated (the pruned surfaces were first wetted with 1/1000 HgCl_2 and after it dried they were painted with Bordeaux paste). (The Bordeaux paste was prepared by mixing 4 oz. of dried precipitate of 4 : 4 : 50 Bordeaux powder with 100 c. c. of raw linseed oil.)
- (vi) Treated with self-boiled lime-sulphur paste in raw linseed oil and then inoculated. (This paste was prepared with 4 oz. of self-boiled lime-sulphur, the proportion of quick lime and sulphur being 2 : 2 mixed in 100 c.c. of raw linseed oil.)

After the paints dried, the treated and untreated portions were inoculated six times with the culture of the fungus No. XXI, covered over with sterilized wet absorbant cotton and then with cellophane bags.

The inoculations were repeated from the middle of April to the middle of August at an interval of a fortnight. After about a month the twigs were examined in the laboratory.

The percentage of infection and the comparison of various treatments are given in Table I. From these tables it is evident that the paste of red lead and copper carbonate in raw linseed oil was the best paint for both mature and immature twigs, to prevent infection of their pruned surfaces.

TABLE I
Comparison of different treatments (1938)

No.	Mean percentage of infection						General mean	Standard error of difference	Critical difference for significance	Whether significant by χ^2 test
	(i) Untreated and uninoculated (control I)	(ii) Untreated and inoculated (control II)	(iii) Red lead and copper carbonate paste	(iv) Red lead and Bordeaux paste	(v) Bordeaux paste	(vi) Self-bolled lime-sulphur paste				
(a) Immature twigs										
1	Per treatment	43.05	86.10	31.04	65.27	74.91	61.55	6.28	13.3185489	Yes $P=0.01$
2	Percentage on general mean	69.90	139.88	51.89	108.04	121.69				
3	Percentage on control I	100.00	200.00	74.19	151.61	174.00				
4	Percentage on control II	50.00	100.00	37.09	75.80	79.03				
(b) Mature twigs										
1	Per treatment	90.27	100.00	31.94	90.27	93.04	83.09	3.77	7.3879492	Yes $P=0.01$
2	Percentage on general mean	108.64	120.35	38.44	108.64	111.97				
3	Percentage on control I	100.00	110.77	35.88	100.00	102.06				
4	Percentage on control II	90.27	100.00	31.94	90.27	93.04				

Dey and Singh [1939] found the paste of red lead with an equal amount of copper carbonate in raw linseed oil to be the best for controlling the stem-brown disease of apple trees caused by *Coniothecium chomatosporum* Corda, which like the stem-brown disease always started from the pruned surfaces.

Thus by using the same paint it is possible to control both the stem-brown and stem-brown diseases of apple trees.

Experiments carried out in 1940

In 1940, another control experiment was started with a view to finding whether lanoline, which is known to hasten the formation of callus, is effective in warding off the disease. Both mature twigs, $\frac{1}{2}$ in. thick, and immature twigs of the apple variety Jonathan were used. Twenty-four twigs, 12 for mature twigs and 12 for immature ones, were selected at random from the orchard. The following six treatments each replicated six times were carried out in each of the 12 trees :—

- (i) Untreated and uninoculated (control I)
- (ii) Treated with lanoline and uninoculated
- (iii) Treated with lanoline and inoculated
- (iv) Untreated and inoculated (control II)
- (v) Treated with lanoline mixed with an equal amount of red lead and copper carbonate and inoculated
- (vi) Treated with red lead and copper carbonate in equal amounts mixed with raw linseed oil and inoculated

After the paints dried the treated and untreated portions were inoculated with the culture of the fungus No. XXI, covered over with wet absorbant cotton and then with cellophane bags. Six inoculations were done after intervals of a fortnight. Twenty days after the last inoculation the twigs were examined in the laboratory.

The mean percentage of infection and the comparison of different treatments are given in Table II.

The following conclusions were arrived at :—

1. Treatment v (a paste of red lead and copper carbonate in equal amounts in lanoline) is significantly better than all the other treatments
2. Lanoline alone (treatments ii and iii) is unable to stop the entry of the fungus
3. Treatments i, ii, iii and iv are not significantly different from one another
4. Treatment vi (a paste of red lead and copper carbonate in raw linseed oil) is significantly better than treatments i, ii, iii and iv :—

$$(a) v > i = ii = iii = iv$$

$$(b) vi > i = ii = iii = iv$$

$$(c) v > vi$$

DISCUSSION

This disease is primarily the disease of currants and was first noticed by Churchill [1891] in the Hudson Valley. In 1896 the currant growers of Marlboro brought the disease to the attention of the New York Agricultural

TABLE II
Comparison of different treatments (1940)

No.	Mean percentage of infection						Whether significant by χ^2 test
	(i)	(H)	(Hi)	(iv)	(v)	(vi)	
	Untreated and inoculated (control I)	Treated with lanoline and urea inoculated	Treated with lanoline and inoculated	Untreated and inoculated (control II)	Treated with lanoline mixed with red lead and copper carbonate in equal amounts and inoculated	Treated with red lead and copper carbonate in equal amounts in raw linseed oil and inoculated	Critical difference for significance
						General mean	Standard error of difference
(a) Immature twigs							
1	Per treatment	98.61	100.00	98.61	38.88	58.71	
2	Percentage on general mean	118.31	120.00	120.00	47.81	72.66	
3	Percentage on control I	100.00	101.42	101.42	39.99	61.41	Yes $P=0.01$
4	Percentage on control II	98.59	100.00	100.00	40.45	62.27	
						82.17	5.53
							$5.53 \times 1.95996 = 10.385788$
(b) Mature twigs							
1	Per treatment	98.61	97.22	100.00	29.08	45.83	
2	Percentage on general mean	126.81	123.23	128.60	37.89	58.93	
3	Percentage on control I	100.00	97.18	101.41	37.91	59.76	Yes $P=0.01$
4	Percentage on control II	98.61	95.83	100.00	29.08	45.85	
						77.76	6.82
							$6.82 \times 1.95996 = 13.3669272$

lation. At this time Durand of Cornell University was investigating a similar currant disease found in western New York [Durand, 1897]. *Nectria cinnabarina* was named as the cause of the disease. In 1899 extensive observations carried out by Grossenbacher and Duggar [1911] established the fact that the currant blight occurring in the Hudson Valley is not caused by *Nectria cinnabarina* but by Fairchild's [1891] 'sterile' fungus. In 1907 a thorough study of the fungus was completed by Grossenbacher and Duggar [1911]. Putterill [1919] described a canker of apple trees in South Africa and named the fungus *Botryosphaeria mali* which differed from the currant fungus, *B. ribis*, in the width of its asci and the size of the stroma. Shear, Stevens and Wilcox [1925] believed that this fungus was not different from *ribis*.

Fenner [1925] described a fruit rot of apples caused by *B. ribis*. Birmingham [1924] described a canker of apple trees in South Wales. In 1934 Smith [1939] described the host range of the fungus and found that it included a list of 34 genera and 20 families of plants. This is a fungus of the type figured and described by Tulasne and Tulasne [1863] as *Dothidea melanops*, later made into the type species of a new genus *Melanops* by Nitschke [1869]. Cesati and Notaris [1863] founded another new genus *Botryosphaeria* in which the forms like *D. melanops* were included, along with some phragmosporic forms. It is claimed, however, that since *Gibberella pulicares* (Fries) Sacc. may have been taken as a type species of *Botryosphaeria*, Nitschke's *Melanops* [1869] should be used for forms like the original *Dothidea melanops* Tul. However, Saccardo [1892] re-defined the limits of the genus *Botryosphaeria* by excluding the forms which develop septate spores. Even though the generic name *Melanops* may replace *Botryosphaeria* for fungi of this type because apparently the type species of the later was subsequently transferred to *Gibberella*, it seems unnecessary to revive an unused name for the isolated case involved in the investigation.

The presence of the two types of pycnidia and one type of perithecia in the life-history of the fungus makes the dissipation of the spores very active in causing primary infections. The comparative measurements of fructification from different hosts by various workers are given in Table III. From these comparative data it will be seen that the Indian strain very closely resembles *Botryosphaeria ribis* of Gross. and Dugg. The slight differences in the measurements of fructifications and spores may have been due to environmental differences.

SUMMARY

The stem-brown disease of apple is caused by *Botryosphaeria ribis* Gross. and Dugg., and usually starts from pruned surfaces and proceeds downwards causing a type of die-back.

Two types of pycnidia are found. Pycnidia of the *Sclerophoma* type are small and the pycnospores are bacillar, allantoid, and do not germinate. The other type of pycnidia are of the *Dothiorella* type, either single or in groups, and the pycnospores are fusoid to oblong, elliptic, hyaline to subhyaline, unicellular and germinate in tap water in 8-12 hours. The perithecia are found rarely in nature. These are either single or in groups of two to eight,

TABLE III

Comparative sizes of fructifications of *Botryosporia ribis* from different hosts in different countries by different workers

Fungus	Host	Locality	Author	Pycnidia (μ)	Pycnospores (μ)	Perithecia (μ)	Asci (μ)	Ascospores (μ)
<i>Botryosporia ribis</i> (Gross. and Dugg.)	Current (<i>Ribis</i> <i>vulgaris</i>)	Hudson Valley, U. S. A.	J. G. Grossenbacher and B. M. Duggar [1911]	175—250 wide.	13—31 \times 4.5 —8	175—250 wide	80—120 \times 17.20	16.23 \times 5—7
<i>Botryosporia mali</i>	Apple (<i>Pyrus</i> <i>mali</i>)	South Africa	V. A. Patterill [1919]	250 \times 190	22.4 \times 4.8	235 \times 142 wide	96 \times 13	19.2—19.5 \times 6.5—8
<i>Botryosporia</i> sp. (Indian strain)	Apple (<i>Pyrus</i> <i>mali</i>)	India, Chambhattia U. P., Kumaun Hills	U. B. Singh [1934]	126.0—294 \times 140—199; Av. 220.7 \times 166.2	9.1—25.9 \times 6.3—7.7; Av. 17.52 \times 6.77	140.0—252 \times 130.2—280; Av. 192.0 \times 190.7	36.4—112.0 \times 44.0—13.9; Av. 77.49 \times 15.39	11.9—28.0 \times 8.4—12.6; Av. 20.1 \times 10.7
<i>Botryosporia ribis</i>	Current (<i>Ribis</i> <i>vulgaris</i>)	From various localities of U. S. A.	N. E. Stevens and A. E. Jenkins [1924]	...	14—31.5 \times 4— 7.5 mostly 18.23 \times 5.7	14.27 \times 5.10 Av. 16.23 \times 6.8

shaped with ostioles. The asci are clavate, eight spored and hyaline. The ascospores are biserial, unicellular, hyaline, fusoid, elliptical to egg-shaped. Paraphyses are present and are filiform. These ascospores germinate readily in tap water in 10-14 hours.

Two isolates of the fungus were taken, one from a single pycnospore, the other from single ascospore. A third foreign strain was also included in the study. The cultural studies of the three strains were carried out on Mapek's agar, potato-dextrose agar and Brown's starch synthetic agar, and it was found that the strain XXI grew better on Brown's starch agar and the strain (St-Br) grew best on potato-dextrose agar. The macroscopic and microscopic observations were also recorded.

All these three strains were found to cause infection of apple twigs through both injured and uninjured surfaces. The progress of infection in all cases was very slow.

One of the three strains was cross-inoculated on cut twigs of pear, apricot, peach and chestnut and was found to infect them.

The cultural studies of the two saltants and the two parents were carried out on three different media—Brown's starch synthetic, cornmeal agar and potato-dextrose agar. The strain St-Br grew best in all three media; one of the saltants XXIa grew better than the parent and other saltant XXIa I, grew best in all the three media.

Pycnospores from the dothiorella type of pycnidia and, to a lesser extent, ascospores carried by water serve as the potent factor for fresh infections. Fresh infections take place from May to July.

A mixture of red lead and copper carbonate in equal amounts in raw linseed oil when applied as a paste on the cut pruned surfaces of apple stem effectively controls the disease.

A paste of red lead and copper carbonate in equal amounts in lanoline is significantly better than a paste of red lead and copper carbonate in equal amounts in raw linseed oil.

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CITRUS ROOTSTOCK TRIALS IN THE PUNJAB

THE VIGOUR OF YOUNG TREES OF SWEET ORANGE, MANDARIN
AND GRAPEFRUIT AS INFLUENCED BY DIFFERENT
ROOTSTOCKS

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(With Plates XV-XVII and one text-figure)

THE most economic and widely accepted method of propagating citrus fruits both in India and elsewhere is by shield-budding a selected scion variety on a suitable rootstock. The success of the combination is markedly affected by both soil and climate, since it is well known that a given rootstock, which is satisfactory in one country or locality, may be a complete failure in another [Brown, 1920 ; Powell, 1930 ; Toxopeus, 1937 ; Webber, 1925]. Furthermore, certain rootstocks are known to exert a considerable influence on the vigour, cropping, etc. of the scion varieties budded or grafted on them [Brown, 1920 ; Bonns and Mertz, 1916 ; Batchelor and Webber, 1939 ; Hatton, 1927 ; 1928-29 ; 1931 ; 1935 ; Hodgson and others, 1937 ; Quinn, 1932 ; Rogers, 1926 ; Rogers and Vyvyan, 1928 ; Singh, 1936 ; Tydeman, 1926-27 ; Webber, 1934 ; Richards, 1938], and certain scions under appropriate conditions show an equally noticeable effect on certain rootstocks [Amos and others 1930 ; Brown, 1920 ; Halma, 1934 ; Hass and Halma, 1929 ; Hatton, 1923 ; 1927 ; Hodgson and others, 1937 ; Roberts, 1929 ; 1931 ; Swarbrick, 1927 ; 1931 ; Tukey and Brase, 1933 ; Webber, 1919 ; Wormald and Grubb, 1924 ; Vyvyan, 1930]. Recent rootstock investigations, especially in England and America, where considerable work has already been carried out on the problems arising directly from the twofold structure of a fruit tree, have tended to emphasize the need for standardized material for all types of investigation. In these countries the growers are at a much greater advantage than their contemporaries living in more backward places, for they will now plant only such scion-stock combinations as have been proved suitable to their conditions.

In our own country, excepting the preliminary work of Brown [1920], Shrivastava [1920] and Prayag [1920], little experimental evidence is available concerning rootstocks for use with the various cultivated species of citrus. Growers have vague and varied impressions of the so-called 'rootstock effect' and it is, therefore, not surprising that all sorts of rootstocks are in common use. The prevalence of low yields, inferior fruit quality and the early decline

of large number of trees is due in a large measure to the promiscuous use of miscellaneous rootstocks by Indian nurserymen.

The available evidence on rootstock investigations points to the fact that the performance of a budded or grafted plant is an expression of the reciprocal effect of the two symbionts. On this basis rootstocks have been designated as vigorous or dwarf, etc. but since the vigour of different scion varieties and species on the same rootstock is known to be different [Hatton, 1935; Roberts, 1929], the terms vigorous and dwarf must be regarded as purely relative. Not only this, the recent findings of various workers [Barker, 1927; Roberts, 1929; Singh, 1936; Tukey and Brase, 1933] show that a scion variety has apparently an inherent growth capacity which it normally exhibits, a capacity which may be dwarfed very considerably but which apparently may not be increased beyond a very small amount. This conclusion leads directly to the suggestion that in all cases, where rootstock influence is a limiting factor to tree size, it is of a dwarfing character. Roberts [1929] goes even as far as to suggest a classification of rootstocks on this basis. According to him the so-called vigorous rootstocks of Hatton [1926] are really neutral since they do not, in any way, limit the free development of the scion. Dwarfing rootstocks, on the other hand, are dominant since they do not allow the scion to develop to its natural capacity.

The present study forms part of a programme of rootstock investigations designed to determine the most suitable rootstocks for sweet orange (Malta), mandarin (Sangtra) and grapefruit in the Punjab, and was carried out during the years 1937-40 upon material planted at Montgomery in February, 1937.

The period of study covers only the vegetative phase of these trees. In view of the fact that growth responses may considerably change during development and maturity, it is essential that the evidence of rootstock influence concerning the two phases of the life of fruit trees should be discussed separately, and it is on this account that the results to date are reported at this stage.

EXPERIMENTAL MATERIAL

Preparation of the material

Nearly all the important varieties of citrus rootstocks were collected from various parts of India and Ceylon, partly to prepare the material for rootstock investigations and partly to make an enquiry into the distribution of different varieties of citrus so as to report on their nomenclature. The latter work will be dealt with in a separate communication.

Seeds as well as cuttings of the rootstock varieties were, as far as possible, obtained from the same parent tree in each case. Both seeds and cuttings were planted at the same time, viz. September 1932. Both the apogamic seedling rootstocks as well as those that could also be propagated by the rooting of stem cuttings were budded over in August 1935, at a uniform height of nearly 9 in. from the ground level, which is the practice commonly followed in the Punjab. It is obvious from the foregoing that the rootstocks, whether raised from seed or by the rooting of stem cuttings, were of uniform age at the time of budding, but it by no means follows that they were of the same vigour. Even within any given rootstock, the progeny differed considerably with respect to height and thickness. However, only fairly uniform

Individuals in each lot were selected for budding and the variants were discarded as advocated by Webber [1920 ; 1932]. As a rule, nearly all the varieties made growth of sufficient vigour to allow of the budding operation, but some rootstocks definitely made much better growth than others.

The experimental material was prepared according to the established nursery practice followed all over the province. Seeds were sown in raised seedbeds in September 1932. The seedlings were dug up in September 1933 and the uniformly vigorous ones only were transplanted in nursery-beds. The stock became fit for budding in September 1935, viz. two years after transplanting, when uniformly vigorous seedlings were budded by a single operator. The budded nursery trees were transplanted in another piece of land at Lyallpur in September 1936 before finally planting the same in the orchard in February 1937 at Montgomery—a place about 200 miles from Lyallpur.

The transplanting of budded trees in the nursery, prior to final planting in the orchard, is not usually practised. But, in this particular case, the operation was considered necessary.

Three scion varieties, namely Malta local (*Citrus sinensis* Osbeck), Sangtra local (*C. nobilis* var. *deliciosa* Swingle) and grapefruit (*C. maxima* var. *Uvacarpa* Merrill and Lee) were budded in August 1935 on to each of the several rootstock varieties propagated from seed and cuttings. Although the preparation of genetically identical material could have been advantageously effected by placing scion buds gathered from a single tree on to carefully selected apogamic seedlings [Webber, 1932 ; Imp. Bur. Fruit Production Tech. Com., 1932], yet clonal vegetatively raised material was also used in order to answer such questions of practical and economic importance as (1) Will the vegetatively propagated rootstocks differ from the apogamic seedlings in their influence on the scion variety notwithstanding the similarity of their genetic build-up ? If so, how and to what extent and degree ? (2) How will the variability in the two sets of material as regards vigour, etc. determined at planting time compare later in the life-history of the experiment ? A further point of particular interest is the utility of the various rootstocks for Malta, Sangtra and grapefruit, under conditions similar to those where the experiments are conducted. This will be determined by investigations into the influence of rootstocks on scions as regards (a) growth and vigour, (b) productivity, (c) fruit quality, (d) resistance to diseases and (e) longevity. Information regarding all these points will be made available when the data collected is sufficient to justify their publication.

Description of the material

As stated before, the experimental material broadly consists of two sets of groups. In one set, the three scion species are budded on to the several rootstocks raised from seed ; in the other set, the same three scion species are budded on to vegetatively propagated rootstocks. With a few exceptions, the rootstock varieties of the experimental material are the same in both the sets. For convenience of reference these sets will be designated as set A and set B. Set A constitutes the three scion-stock groups prepared by budding three scion species on several rootstocks propagated from seed, and set B includes the other three groups prepared by budding the same three

scion species on the rootstocks that were propagated by vegetative means. The specifying numbers allotted to the rootstocks in both sets of groups are purely arbitrary and are not based on any systematic study.

The various scion-stock combinations in sets A and B are tabulated in Table I.

TABLE I
Different scion-stock combinations under experiment

Name of scion-stock combination				No. of trees under study	
Set A.— (i) Malta local on rootstock No. 43 . . .				24	} 120
	Ditto	21 . . .	24		
	Ditto	20 . . .	24		
	Ditto	50 . . .	24		
	Ditto	9 . . .	24		
(ii) Sangtra local on rootstock No. 43 . . .				18	} 90
	Ditto	9 . . .	18		
	Ditto	21 . . .	18		
	Ditto	20 . . .	18		
	Ditto	50 . . .	18		
(iii) Grapefruit on rootstock No. 43 . . .				24	} 120
	Ditto	20 . . .	24		
	Ditto	47 . . .	24		
	Ditto	50 . . .	24		
	Ditto	9 . . .	24		
Set B.— (i) Malta local on rootstock No. 20 . . .				18	} 72
	Ditto	43 . . .	18		
	Ditto	50 . . .	18		
	Ditto	9 . . .	18		
(ii) Sangtra local on rootstock No. 43 . . .				18	} 72
	Ditto	20 . . .	18		
	Ditto	9 . . .	18		
	Ditto	50 . . .	18		
(iii) Grapefruit on rootstock No. 43 . . .				18	} 72
	Ditto	20 . . .	18		
	Ditto	50 . . .	18		
	Ditto	9 . . .	18		

The material in sets A and B is planted separately in contiguous fields but the lay-out, viz. arrangement of plots, position of main water channels, block water channels and position of paths, etc. is uniform in each case.

LAY-OUT

It is evident from the foregoing section that sets A and B have each three different groups of scion-stock combinations, depending upon the number of scion varieties employed. There are thus in all six different groups of

experimental material planted in six different fields. The randomized block method, being most adaptable to field experiments in horticulture, is the one here adopted. The arrangement and distribution of different scion-stock combinations within the plots is explained by Fig. 1. The method of lay-out of each field is the same as shown in this figure.

A glance at the lay-out, given in Fig. 1, shows that there are seven rootstocks under trial in set A, of which a comparative study of only five is reported in this paper. The position of the trees on rootstocks 44 and 1, which have not been dealt with, is shown by shaded sub-plots in the figure. These two rootstocks have turned out to be horticulturally the same as No. 20 (rough lemon). Thus, rough lemon is represented thrice in each replicate, whereas each of the four remaining rootstocks is represented only once in each block. With a view to keeping uniformity in the number of replications (one sub-plot to represent each treatment in each block) it was necessary to select one out of the three identical varieties originally planted; No. 20 was selected because in set B, rootstocks 44 and 1 were not included for trial and only 20 was used.

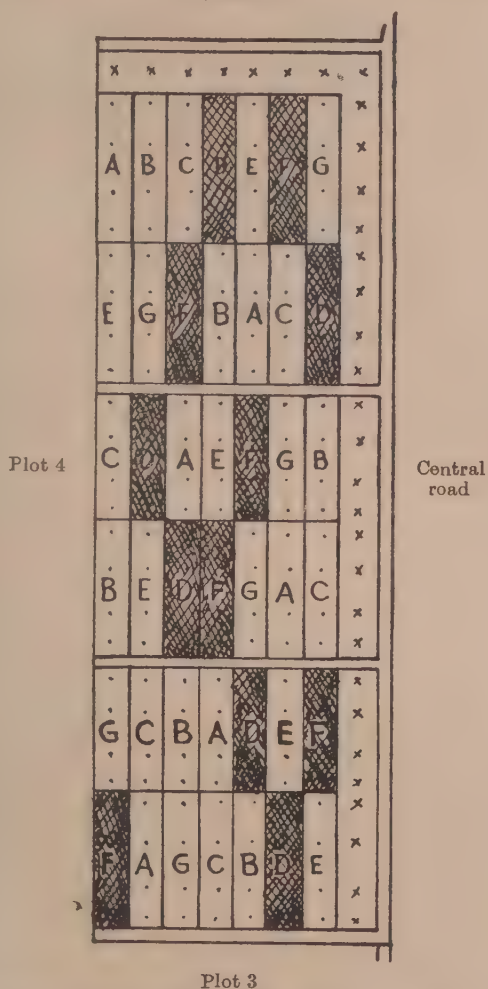


FIG. 1. The distribution of seven scion-stock combinations in six blocks in plot No. 5

- | | |
|-------------------------------------|-----------------------------------|
| A. Malta local on rootstock No. 20* | B. Malta local on rootstock No. 9 |
| C. " " " No. 21 | D. " " " No. 1* |
| E. " " " No. 43 | F. " " " No. 44* |
| G. Malta local on rootstock No. 50 | |

..... Experimental trees × × × × × Non-experimental trees
 ===== Water channels

*Rootstocks No. 20, 1 and 44 are found to be one and the same.

The piece of land for each experiment is divided into six blocks of similar dimensions. Thus, six replications are provided for each series of scion-stock combination. Each block in turn is divided into as many plots of equal size and shape as the number of rootstock varieties to be compared. The allocation of scion-stock combinations to particular plots was determined by drawing lots, as a result of which each combination is represented once in each block. The dimensions of the plots vary from 20 ft. \times 60 ft. to 20 ft. \times 80 ft. depending upon the number of trees planted. In other words, a unit of three to four trees of any particular scion-stock combination is replicated six times, so that there are 18-24 trees of this in any one experiment.

The planting of all the six experimental fields follows the square system. A uniform distance of 20 feet from tree to tree and row to row is being adopted.

Due attention was given to the variability within the experimental material at the time of planting. The trees within each lot were roughly graded according to vigour and planted in such a way as to include only fairly uniform ones of all combinations in each block. There being six blocks placed end to end in each case (Fig. 1), the variability within the material is allowed for in block variance when computing the data.

METHODS EMPLOYED

Soon after planting the material in 1937, trunk circumference measurements of all the trees were taken at a uniform height of approximately one foot from the ground level. The places of measurement were marked with white paint so that the girth measurements could be taken at a fixed point year by year. The union of the scion with the rootstock was invariably below the place of measurement.

The girth measurements were taken again in 1938, 1939 and 1940 at yearly intervals with a view to determining the growth response during early years of the various scions growing on different rootstocks. The girth measurements serve as a measure of the vigour of the trees in these trials. The data regarding girth, for all the years, have been examined statistically by the analysis of variance [Fisher, 1934]. The differences above the 5 per cent level only are taken as significant. On the basis of the information thus gathered the various scion-stock combinations have been grouped so as to show at a glance their behaviour from year to year.

DISTINGUISHING NAMES OF THE ROOTSTOCK VARIETIES EMPLOYED

The morphology and other characters of the rootstock varieties employed in this study along with others in the collection have been studied during the last two years in order to clarify their nomenclature. This work, when completed, will be published separately, but in the meantime the rootstocks under trial have been properly identified as follows:—

TABLE II

Distinguishing names of the rootstock varieties under trial

Names under which received	Arbitrary specifying numbers allotted as referred to in this paper	Popular local names in the Punjab	English equivalents	Specific names
Mitha . .	50	Mitha .	Sweet lime .	<i>C. aurantifolia</i> var. Swingle
Khatti . .	20	Jatti khatti	Rough lemon	<i>C. limonia</i> Osbeck
Chakotra .	47	Chakotra .	Shaddock .	<i>C. maxima</i> Merrill
Puranj .	9	Mokari .	Citron . .	<i>C. medica</i> Linn.
Kharna khatta	43	Kharna khatta	Nil	<i>C. karna</i> Raf.
Nasnaran .	21	Nil	Nil	<i>C. japonica</i> Thumb

Beside the six rootstocks mentioned above, another apparently distinct species, viz. *khatta* (*C. aurantium* Linn.) was also included, but on studying morphological characters it was found to be really rough lemon. It is evident that the material used as rootstocks does not include species like *C. aurantium*, *C. nobilis* and *C. sinensis*, etc. These species, especially the first one, are probably indigenous to India, and may, therefore, have great possibilities as rootstocks in view of the fact that they are already in use in some of the citrus-growing countries of the world [Coit, 1927; Hume, 1930; Howell, 1930]. In preparing further material, these species will be included as well as others that are now growing in the rootstock collection plot at Montgomery. Furthermore, certain points of practical importance regarding these rootstocks are being studied in preparation to a further and more extended trial of rootstock material.

PRESENTATION OF RESULTS

As mentioned in previous sections, the girth measurements of all the trees constituting the various scion-stock combinations were taken annually (1937-40) as an index of their vigour as influenced by the rootstocks on which they were stem-budded. The data for 1937 relates to the behaviour of maidens at the time of planting in the orchard, that for 1938 as the behaviour of one-year old plants and that for 1940 as the behaviour of three-year old plants. The material constitutes two sets, set A and set B. In set A apogamic seedlings have been used as rootstocks and in set B the rootstocks used were propagated by the rooting of stem cuttings. The genetic composition of any particular rootstock in both the sets is thus identical but apogamic seedlings

and rooted cuttings being physiologically different, the results of their performance with different scions are presented separately as under :—

Scion-stock combinations of set A with different scion varieties

1. *Malta local*.—The mean data and significant difference at the per cent level are compiled in Table III which shows the relative size of the year old plants of this variety as influenced by different rootstocks (Pls XV-XVII).

TABLE III

Performance of Malta local variety with certain rootstocks of apogamic origin

Year	Average circumferential measurements in cm. for rootstock numbers					General Result	S. E.	Significance $P = 0.01$
	43	21	20	50	3			
1937 . . .	5.08	5.63	3.55	3.35	3.61	4.25	0.23	0.7
1938 . . .	5.88	6.25	4.25	4.06	3.96	4.45	0.29	0.8
1939 . . .	9.97	8.83	8.50	7.73	6.11	8.23	0.57	1.8
1940 . . .	16.7	15.7	15.4	14.0	13.2	14.97	0.66	2.1

The performance of different scion-stock combinations has first to be examined separately for all the four years before discussing them together. The resultant effect of the various combinations on the vigour of the scion is tested by comparing the difference between average circumferential measurement figures relating to any pair of combinations with the significant difference figure given in the last column for each year. On the basis of this comparison, the various rootstocks, differentiating themselves from one another, are put in different groups. When certain rootstocks are found to exhibit only small differences as compared with the significant limit, they are put in one and the same group. The grouping of the various rootstocks, based on this statistical examination of the data given in Table III, is made as under :—

Year	Group 1	Group 2
1937 . . .	21; 43	9; 20; 50
1938 . . .	21; 43	20; 50; 9
1939 . . .	43; 21; 20	50; 9
1940 . . .	43; 21; 20	50; 9

The figures show that the above five rootstock varieties fall into two groups even in the nursery stage. Thus, rootstocks 21 and 43 are associated with the more vigorous trees of Malta local variety, whereas the remaining rootstocks (in group 2) are associated with trees of medium vigour. The grouping of the rootstocks for 1938, viz. one year after planting, was substantially the same except in their order of vigour within group 2. By the end of the second year of their life in the orchard, i.e. in 1939, the various rootstocks still fall into two groups but their order of vigour is considerably changed. Thus, rootstock 20 is now bracketed with 43 and 21 as being vigorous, and tree



FIG. 1. Local Malta on *kharna khatla*



FIG. 2. Local Malta on *nasmaran*



FIG. 2. Local Malta on *miha*



FIG. 1. Local Malta on *jati khatti*

ck No. 43 are now slightly more vigorous than those on No. 21. Rootstocks 50 and 9, however, remained in group 2. The behaviour of the different rootstocks in 1940 remained exactly the same as during 1939 and it appears that rootstocks 50 and 9 may ultimately prove to be dwarfing for the Malta variety.

Sangtra local.—The mean data and significant difference at the 5 per cent level are compiled in Table IV, which shows the relative size of three year old trees of this variety as influenced by different rootstocks.

TABLE IV

Performance of *Sangtra local* variety with certain rootstocks of apogamic origin

Year	Average circumferential measurements in cm. for rootstock numbers					General mean	S. E.	Significant difference for $P=0.05$
	43	9	21	20	50			
1937	4.97	3.42	3.19	2.17	2.33	3.22	0.18	0.55
1938	6.14	4.53	4.16	2.83	3.38	4.21	0.17	0.53
1939	11.77	9.34	8.61	7.26	7.96	8.99	0.30	0.94
1940	19.5	16.1	15.9	15.5	14.5	16.28	0.41	1.32

The grouping of the various rootstocks, based on the statistical examination of the data given in Table IV above, is made as under :—

Year	Group 1	Group 2	Group 3	Group 4
1937	43	9; 21	50; 20	..
1938	43	9; 21	50	20
1939	43	9; 21	50; 20	..
1940	43	9; 21; 20	50	..

The above grouping shows that with this variety rootstock No. 43 (*kharna*) is differentiated from the rest in that the trees of the *Sangtra local* variety are extremely vigorous even from their nursery stage and that this has been maintained throughout the period covered by this report. The order of vigour are trees on rootstocks 9 and 21— an order which remained unchanged for all the four years. By 1940, however, one more rootstock, viz. No. 20, has jumped up in this group. In the third and fourth years the various rootstocks did not materially change their order of vigour. Rootstock 50 alone remained in the third group. Rootstock 50 may, therefore, ultimately prove to be a dwarfing rootstock for *Sangtra local* scion variety.

Grapefruit (Marsh Seedless).—The mean data and significant difference at the 5 per cent level are compiled in Table V, which shows the relative size of three-year old trees of this variety as influenced by different root-

TABLE V

Performance of grapefruit (Marsh Seedless variety) with certain rootstocks of apogamic origin

Year	Average circumferential measurements in cm. for rootstock numbers					General mean	S. E.	Significant difference $P=0.05$
	43	20	47	50	9			
1937 . . .	3.99	3.72	3.75	3.19	2.82	3.50	0.158	0.5
1938 . . .	5.43	5.43	5.02	4.42	4.25	4.91	0.208	0.6
1939 . . .	10.36	10.18	9.27	8.06	8.30	9.18	0.335	1.0
1940 . . .	18.7	18.3	16.6	14.7	12.4	16.12	0.477	1.5

The grouping of the various rootstocks, based on the statistical examination of the data given in Table V above, is made as under :—

Year	Group 1	Group 2	Group 3	Group 4
1937. . .	43 ; 47 ; 20	50 ; 9
1938. . .	43 ; 20 ; 47	50 ; 9
1939. . .	43 ; 20 ; 47	9 ; 50
1940. . .	43 ; 20	47	50	9

It is evident that, in 1937, with grapefruit as the scion variety, the three rootstocks Nos. 43, 47 and 20 are associated with more vigorous trees while the remaining two, viz. 50 and 9, formed a class which proved to be less vigorous. The relative performance of the three vigorous rootstocks did not change in the following year except that the order of vigour of stocks 47 and 20 in group 1 is reversed. A year later, viz. the beginning of 1939, the order of vigour of rootstocks in group 1 still remained unchanged, although a slight change occurred in group 2. By the beginning of 1940, however, the five rootstocks under trial differentiated into four different groups with regard to their influence on the vigour of grapefruit scion. Thus, grapefruit trees on stocks 43 and 20 are of greatest vigour, those on 47 are of medium vigour, while those on 50 and 9 are dwarf notwithstanding the fact that No. 9 is significantly more dwarfing than No. 50.

Scion-stock combinations of set B with different scion varieties

1. *Malta local*.—The mean data and significant difference at the 5 per cent level are compiled in Table VI, which shows the relative size of the 5 year old trees of this variety as influenced by different rootstocks.



Local Malta on *mokari*

TABLE VI

Performance of Malta local variety with certain rootstocks raised by the rooting of stem cuttings

Year	Average circumferential measurements in cm. for rootstock numbers				General mean	S. E.	Significant difference for $P=0.05$
	20	43	50	9			
1937	4.08	4.65	5.36	3.60	4.38	0.357	1.12
1938	6.09	6.09	6.66	4.74	5.89	0.30	0.94
1939	11.49	11.39	10.63	8.36	10.47	0.36	1.14
1940	19.1	19.0	16.4	13.1	16.91	0.46	1.47

The grouping of the various rootstocks, based on the statistical examination of the data given in Table VI, is made as follows:—

Year	Group 1	Group 2	Group 3
1937	50; 43	20; 9	..
1938	50; 43; 20	9	..
1939	20; 43; 50	9	..
1940	20; 43	50	9

The grouping of rootstocks shows that at the nursery stage in 1937, they fell into two groups only. In group 1 occur the rootstocks 50 and 43 which were associated with more vigorous trees of the Malta local scion. The trees of rootstocks 20 and 9 form a less vigorous class. One year after planting, in 1938, rootstock 20 had moved from group 2 into group 1, but the order of vigour of the two rootstocks in group 1 did not change. In 1939, the order of vigour of rootstocks in group 1 changed considerably. Thus, rootstock 50, which occupied 1st position in 1937 and 1938, went to the 3rd position and rootstock 20, which occupied last position in 1938, came to the 1st position. However, one should not lose sight of the fact that, despite the change in the order of vigour of these rootstocks, they are statistically identical, but this abrupt behaviour of stock No. 50 promises further segregation among the varieties in this group. Rootstock No. 9, however, remained in group 2 as before. In 1940, rootstock 50 dropped back to group 2 but the dwarfing two stocks in group 1 maintained their performance. It cannot be foretold which of the two rootstocks in group 1 will ultimately prove to be the most vigorous, but it would appear that stock No. 9 is likely to prove dwarfing stock for this scion variety more so than any other, and that stock No. 50 may also prove dwarfing to this variety.

2. *Sangtra local*.—The mean data and significant difference at the 5 per cent level showing the relative size of three-year old trees of this variety influenced by different rootstocks are compiled in Table VII.

TABLE VII

Performance of Sangtra local variety with certain rootstocks raised by the rooting of stem cuttings

Year	Average circumferential measurement in cm. for rootstock numbers				General mean	S. E.	Significant difference for $P=0.05$
	43	20	9	50			
1937	5.76	2.39	2.48	4.23	3.72	0.301	0.95
1938	7.09	4.20	4.07	5.78	5.29	0.468	1.48
1939	13.24	9.71	10.07	11.02	10.99	0.335	1.06
1940	21.1	16.9	16.8	16.6	17.8	0.48	1.5

The grouping of various rootstocks, based on the statistical examination of the data given in Table VII, is made as follows :—

Year	Group 1	Group 2	Group 3
1937	43	50	9; 20
1938	43; 50	..	20; 9
1939	43	50; 9	20
1940	43	20; 9; 50	..

The data show that throughout the four-year period, trees of Sangtra scion variety on rootstock No. 43 are more vigorous than on the remaining three rootstocks. Rootstock No. 50, which proved next in order of vigour during the first three years, had, by the end of the fourth year, proved a dwarfing rootstock. No. 20, on the other hand, which was dwarfing as compared with others during the first three years, was eventually shown to have an invigorating effect. At the end of four years, Nos. 20, 9 and 50 are not significantly different from one another and, as a class, they are significantly less vigorous than No. 43.

3. *Grapefruit (Marsh Seedless)*.—The mean data at the 5 per cent level are compiled in Table VIII which gives the relative size of three-year old trees of this variety as influenced by different rootstocks.

TABLE VIII

Performance of grapefruit (Marsh Seedless) with certain rootstocks raised by the rooting of stem cuttings

Year	Average circumferential measurements in cm. for rootstock numbers				General mean	S. E.	Significant difference for $P=0.05$
	43	20	50	9			
1937	6.11	3.65	4.88	4.15	4.70	0.324	1.03
1938	7.75	5.50	5.71	5.61	6.13	0.380	1.21
1939	12.94	11.39	10.04	9.77	11.03	0.4	1.28
1940	21.2	20.3	15.8	15.5	18.23	0.375	1.2

The grouping of various rootstocks, based on the statistical examination of the data given in Table VIII, is made as follows :—

Year	Group 1	Group 2	Group 3
1937	43; 50	9; 20	..
1938	43	50; 9; 20	..
1939	43	20	50; 9
1940	43; 20	..	50; 9

At the time of planting, i.e. at one-year old, the rootstocks 43 and 50 were both associated with vigorous trees of the Marsh Seedless grapefruit variety, while the remaining rootstocks, viz. 9 and 20, formed a second class which differed from them in this respect. By the end of the next year, rootstock No. 43 alone remained in the vigorous group, and the remaining tree fell into group 2. In 1939, viz. two years after planting in the orchard, further differentiation took place in the rootstocks of group 2, by which trees on No. 20 became significantly more vigorous than those on either of No. 50 or 9. By 1940, trees on rootstock No. 20 became still more vigorous, and they are now nearly as vigorous as those on rootstock No. 43, whereas rootstocks No. 50 and 9 appear to be dwarfing ones.

DISCUSSION

Rootstock No. 43

The performance of rootstock No. 43 [*Kharna khatta* (C. karna Raf.) synonyms : *Id lemon* (Poona, Bombay), *Soh sarakar* (Assam), *Mokari* (Renala Khurd, Punjab) is outstanding in that it is associated with the most vigorous trees of all the three scion varieties budded on it. This is true for both seedling and cutting material. It differentiates, in this respect, from most of the remaining rootstocks as early as one year after budding. The vigorous growths of trees on this rootstock, coupled with the fact that its seedlings are usually ready for budding earlier than those of other varieties, indicate that, from the nurseryman's standpoint, it is a rootstock which may become widely used because, other things being equal, good nursery trees of a given scion variety can be produced relatively early. Furthermore, the consistent behaviour of scions on this rootstock for the first four years after budding strengthens the view that, as compared with other varieties included in these trials, this one may ultimately produce trees of the largest size. This point is of special significance in so far as it holds good for all the three scion varieties employed.

Brown [1920] reported a similar invigorating influence of *kharna* rootstock which, according to the photograph of fruit in Plate VI, fig. 1 presented by him, appears to be of the same variety as *kharna khatta* employed in the present investigations. Brown, however, translated this as rough lemon, a variety from which it differs in almost every respect. It would appear, therefore, that Brown did not employ the true rough lemon in his trials, although he has reported the results of *kharna khatta* as being due to rough lemon. The results of Brown have also been quoted by subsequent workers [Gardner *et al.*, 1922] as the effects of rough lemon rootstock upon scion growth, a repetition of which should be avoided.

So far as the present authors are aware, *kharna khatta* as a rootstock has not been employed in other citrus-growing countries of the world, and it is very probable that this species is a native of India. Its performance in these trials warrants an extension of its use on an experimental scale in this country and elsewhere.

2. Rootstock No. 20

The use of this rootstock [*jatti khatti* or rough lemon (*C. limonia* Osbeck synonyms: *Khattu* (Renala Khurd and Lahore, Punjab) *Khatti* (Lyalpur and Shahdara, Punjab)] has been investigated in almost every citrus-growing country of the world. It should, however, be clearly understood that this rootstock was not employed by Brown [1920]; Shrivastava [1920] and Prayag [1920] in their investigations in North-West Frontier Province, Central Provinces and Bombay and that *Id lemon* and *jamburi* said to be the varieties of rough lemon [Imp. Bur. Fruit Production, Tech. Com., 1932] have nothing in common with the real rough lemon of South Africa, California and Florida, etc. [Coit, 1927; Hume, 1930; Powell, 1930]. It follows, therefore, that no work has yet been done anywhere in India to elucidate the influence of rough lemon on the commercially grown citrus scion varieties of this country.

As a result of the satisfactory growth of trees on rough lemon in most parts of the citrus world, it is the one most used in South Africa [Marloth, 1938; Powell, 1930]. In Arizona, California and Java [Coit, 1927; Marloth, 1938; Toxopeus, 1936] it is used to a limited extent. In Florida [Batchelor and Webber, 1939; Coit, 1927; Hume, 1930; Marloth, 1938] it is the main rootstock for very sandy soils although, according to Davis [1928], it thrives equally well on heavy loams. In Dominica, rough lemon has proved to be a good stock for acid limes [Imp. Bur. Fruit Product. Tech. Com., 1932]. In Australia [Marloth, 1938] it is employed extensively, and in the Punjab it is also one of the widely used rootstock varieties. Cheema [1929] advises rough lemon as the most suitable rootstock for lemons in western India, though the species actually used is different from real rough lemon. It is evident from the above, therefore, that with rough lemon the problem appears to be one of adaptability to local soil and climatic conditions rather than congeniality between stock and scion, since cases of absolute failure or imperfect union have not been reported from anywhere.

The data presented in the foregoing section show that rough lemon is a promising rootstock and its growth influence in case of the sweet orange (Malta) and grapefruit scions may ultimately be in line with *kharna khatta*, but in the case of Sangtra scion variety it may not prove equally vigorous. It is, however, significant that even with sweet orange and grapefruit scions, it did not produce as vigorous nursery trees in the same period as was done by some other rootstocks. All the same, the experience of workers in other countries [Coit, 1927; Hume, 1930], that this rootstock is associated with vigorous vegetative growth of young trees, is also corroborated by the data here reported.

3. Rootstock No. 50

Sweet lime [*mitha* (*C. aurantifolia* var. Swingle)] is, perhaps, indigenous to India. On its own roots, it is quite vigorous and spreading but when used

as a rootstock for sweet orange, mandarin and grapefruit, the resulting trees are dwarfed. It is a favourite with nurserymen throughout the Punjab, partly due to the prevalent belief that it improves the texture and quality of fruit of the scion varieties worked on it, and partly to its ability to root readily from stem cuttings producing well grown nursery stock in a comparatively short period. The latter point concerns mainly the nurseryman, who is chiefly interested in the production of plants in a short space of time, but the former point concerns the grower and the fruit industry in general and is, therefore, more worthy of consideration and experimentation.

The data show conclusively that, with all the three varieties in set A, sweet lime rootstock behaves as a dwarfing type. In set B, however, the trees on this stock are vigorous when young, but by the beginning of 1940, the trees on some other rootstocks had significantly outgrown them. The difference in the vigour of scion varieties in sets A and B, brought about by this rootstock for the first few years was, therefore, of a transitory nature and was mainly due to the increased vigour of nursery stock in set B at the time of budding. Evidently therefore the nursery stock of sweet lime, raised by the rooting of cuttings, became fit for budding comparatively much earlier than that raised from seed. Not only this, with the exception of *galgal* (hull lemon) which is not employed in these trials, sweet lime cuttings resulted in nursery stock of a better size and vigour than that of the remaining stock varieties under trial in set B. Since time of budding was the same in sets A and B, the stocks in set B made much better growth than those in set A and even within set B the sweet lime stock made better growth than others in view of its better rooting capacity and was consequently at an advantage to induce better growth of scion buds placed in it. This initial advantage, however, was not maintained and this rootstock must now be regarded as dwarfing in character as in the case of set A. All the three scion varieties worked on sweet lime in sets A and B are somewhat dwarfed, and these effects are in general agreement with the results obtained by Brown [1920] in India and are also in accord with the experience of growers in south eastern Mediterranean countries, especially in Palestine (Hodgson, 1931; Powell, 1930), where Jaffa or Shamouti variety of sweet orange is mainly grown on sweet lime.

4. Rootstock No. 21

According to Bonavia [1880] this rootstock, [*mosaicum* (C. japonica Thumb)] resembles in character the *reshmi* orange of the United Provinces, India, and is not a true mandarin although it is called mandarin in Ceylon. It was introduced into the Punjab through the courtesy of the Curator, Royal Botanic Gardens, Peradeniya, Ceylon, and was included only in set A (rootstocks of apogamic origin) as the cuttings did not root well. On its own roots, this rootstock is comparatively dwarf, but when used as a rootstock for sweet orange (Malta local) it has produced maindens of greater vigour than those of the most vigorous known rootstocks. On transplanting to the orchard, the trees on this rootstock naturally received the greatest setback and some of them even died, but the remaining ones are now growing well. On the other hand, when used as a rootstock for mandarin (Sangtra) it produced maindens of medium vigour only, which when transplanted grew very well indeed. Furthermore, the results (Tables III and IV) show that, both with sweet orange and mandarin as

the scion varieties, this rootstock maintained its nursery performance, viz. that of producing vigorous trees in case of the former and dwarf trees in case of the latter throughout the course of these investigations. This differential response may be partly correlated with the fact that the union of this rootstock with Malta scion is perfect, but with Sangtra scion the stock stem outgrows the scion stem.

5. Rootstock No. 9

This species [*mokari* or citron (*Citrus medica* Linn.), synonyms : *Turanj* (Renala Khurd, Punjab), *Sak limboo* (Poona), *Nattaran* (Ceylon), *Sohmad* (Assam)] is a native of India [Powell, 1930]. It has two forms : one where the rind is very rough, corrugated and is orange on ripening (C. Mahalung, Poona) and the other where the rind is smooth and is yellow on ripening. It is the latter form that has been employed in the trials reported here.

The nursery performance of this species is equal to that exhibited by our best rootstocks. It is easily grown from cuttings and both cutting and seedling stocks reach the budding stage as quickly as those of any other stock. Probably the greatest point in its favour is the fact that variability in the seedbed with regard to the height and vigour of seedlings is low and in consequence only a small percentage of seedlings has to be discarded at the transplanting time. The main disadvantage appears to be its extreme susceptibility to citrus canker and gum diseases.

Out of the three scion varieties, viz. sweet orange (Malta local), mandarin (Sangtra local) and grapefruit, with which it has been tried as a rootstock, it has given good union only with mandarin while in the remaining two cases the scion portion has invariably outgrown the rootstock.

The data show conclusively that with sweet orange and grapefruit, where it is not quite compatible, its effect is that of dwarfing nature but in case of Sangtra local scion the trees on this rootstock are fairly vigorous. In this respect, therefore, like *nasnaran* it has responded differently with different scion varieties. It has been largely used as a rootstock in Egypt [Brown, 1936] with the result that trees budded or grafted on it remain inferior in vigour and resistance to diseases. This experience of Egyptian growers is in conformity with the results obtained in the Punjab.

6. Rootstock No. 47

As far as could be ascertained, shaddock [*chakotra* (*Citrus maxima* Merrill)] has never been employed as a rootstock in the Punjab for orange, mandarin and grapefruit. However, in view of the remarks of Webber [1925] that shaddock has been favourably commented on as a rootstock for grapefruit in South Africa, it was considered desirable to include this in the citrus rootstock trials, especially in the case of grapefruit.

It has made a good union with grapefruit and the results show that trees on this stock are fairly vigorous. Although the trees have only completed three years in the orchard, some of them growing on this rootstock are severely affected with mottle leaf. Whether it is purely accidental that the portions of the land, on which the trees of this scion-stock combination are planted, are deficient in certain elements causing mottle leaf or whether this rootstock itself is responsible for this malady cannot at present be determined.

SUMMARY

The influence of certain rootstocks on the vigour of young stem-budded scions of sweet orange (Malta), mandarin (Sangtra) and grapefruit has been studied at Montgomery, Punjab, for the years 1937-40.

The results show that:—

- (a) *Kharna khatta* (*C. karna* Raf.) is associated with the most vigorous trees irrespective of the scion variety budded on it.
- (b) *Jatti khatti* or rough lemon (*C. limonia* Osbeck) is also associated with vigorous-growing trees, but it does not equal *kharna khatta* in this respect.
- (c) *Mitha* or sweet lime (*C. aurantifolia* var. Swingle) appears to be a dwarfing rootstock for each of the three scion species budded on it.
- (d) *Nasnaran* (*C. japonica* Thumb) and *mokari* or citron (*C. medica* Linn.) both appear to be variable in their influence as rootstocks. For instance, sweet orange on *nasnaran* has proved vigorous, but mandarin trees on this rootstock are decidedly dwarfed. Similarly, plants of sweet orange and grapefruit are dwarf on *mokari*, but those of mandarin on the same rootstock are vigorous.
- (e) Grapefruit trees on *chakotra* or shaddock (*C. maxima* Merrill) show mottle leaf but it has not yet been determined whether this is due to soil or to rootstock.

The vigour of different scion species has been shown to be variable on one and the same rootstock in case of two rootstock species, namely *nasnaran* and *mokari*. The terms vigorous and dwarf usually assigned to rootstocks should be used only in connection with specified scion varieties.

The vigour of an unworked rootstock is no criterion of its vigour as a rootstock when grafted with certain scion varieties. For instance, *nasnaran* which is comparatively dwarf on its own roots has, when used as a rootstock for sweet orange, given rise to trees equal in vigour to those on the so-called vigorous rootstocks. *Mitha* (sweet lime), on the other hand, is quite vigorous and spreading on its own roots but, as a rootstock, it has dwarfed all the scion varieties budded on it in spite of its having made good union with the scion varieties. *Mokari* (citron), which is fairly dwarf on its own roots, has produced Sangtra plants of good vigour.

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UTILIZATION OF VIRGINIA TOBACCO SEED IN THE MADRAS PROVINCE

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THERE is an area of about 3,00,000 acres under tobacco in the Madras province, of which nearly 1,50,000 acres is confined to the Guntur district 50,000 acres to the Kistna and Godavari districts and the rest to the other parts of the province. Out of this, about 1,25,000 acres are grown with Virginia tobacco. Nearly 80 per cent of this area, in turn, is in the Guntur district and the rest in the Kistna and Godavari districts along the borders of the two rivers. The crop is allowed to flower and pod freely—unlike *natu*, the local cigar variety, which is invariably topped—in nearly 90 per cent of the Virginia tobacco-growing area to impart proper texture, body and colour to the leaf and make it fit for cigarette manufacture.

Seed, its oil content and general properties

An average crop of Virginia tobacco yields about 150-200 lb seed per acre. After the leaf picking is over and before the crop is pulled, sheep and goats are allowed to eat the pods freely when ripe, and they seem to relish it and do well. Majority of the seed pods are, however, burnt along with stalks which at present form the main fuel in use in all Virginia tobacco-growing areas. From preliminary trials it was found that the seed contained some oil and hence it was thought desirable to find some use for it. On extraction by the cold-drawn process in the country wooden mill (*chekku*) and by the hot-drawn process generally adopted for extracting castor oil, oil to an extent of 25-30 per cent could be got. The cold-drawn oil was thin, transparent, light yellow, pleasant smelling, agreeable to taste, and almost identical with first class gingelly oil. The hot-drawn oil was thin, slightly turbid yellow, pleasant smelling, and slightly bitter in taste. The seed and cake were readily eaten by sheep, goats and cattle. Viands made out of the cold-drawn oil obtained from well-washed seeds were not different from those made out of gingelly oil. Those prepared in the hot-drawn oil and in cold-drawn oil extracted from unwashed seeds were rather bitter to taste. The extraction of oil by the cold-drawn process as is done in the case of gingelly seeds will prove a good cottage industry in the Virginia tobacco areas, but there are not enough country wooden oil mills available in the areas for the purpose. Extraction by the hot-drawn process in the hand presses used for extracting castor oil is quite possible. Tobacco seeds, however, seem to

and a higher temperature and more pressure than castor seed for efficient extraction and to achieve this some structural changes in the oil presses now in use may be necessary. When the seed is pressed by this process, it should be crushed in flour mills before pressing for efficient extraction of oil.

Chemical investigations of seed oil and cake

Samples of oil and cake from the same batch were sent to several analysts in India and the results supplied by them are incorporated in Tables I and II. For purposes of comparison, the results obtained by investigators in other parts of the world on the subject are produced in Tables IV and V.

TABLE I

Results of analysis of Guntur Virginia tobacco seed for food and mineral values

(By the Government Agricultural Chemist, Coimbatore)

Head of analysis	Per cent
<i>a. Food values—</i>	
Moisture	6.05
Ash	3.76
Crude proteins ($N \times 6.25$)	23.88
Ether extractives	35.77
Crude fibre	16.77
Carbohydrates (by difference)	13.77
Total	100.00
<i>Mineral values—</i>	
Insolubles	0.31
Albuminoids (true proteins)	22.80
Nicotine	Absent*
Lime (CaO)	0.21
Potash (K_2O)	0.94
Phosphoric acid (P_2O_5)	1.09
Nitrogen (N)	3.82

* As observed by the Imperial Agricultural Chemist, New Delhi

The results obtained and the conclusions drawn therefrom were in general conformity with those obtained elsewhere and establish the fact that Guntur Virginia tobacco seed contains about 35-37 per cent of oil, is free from nicotine and can be used as an edible or semi-drying oil and for soap making or for illumination purposes. Its uses as an edible oil in Bulgaria are reported by Jamieson [1932] and as a semi-drying oil by Paris [1920] and Pyat-nitz [1929]. Samples of hard and soft soaps, prepared at the Kerala Soap Institute, Calicut indicate that the oil can be used for this purpose with success.

The cake contains about 30-35 per cent crude proteins, 29-34 per cent albuminoids, 16-17 per cent ether extractives, about 27 per cent carbohydrates, and is free from nicotine. It compares favourably in its food values with gingelly cake so extensively used for cattle feed in this province (Table VI) and cattle eat the cake freely. Feeding trials substituting tobacco seed cake for groundnut cake were conducted over a period of five weeks at the Agricultural Research Station, Guntur. The feeding did not bring about any adverse effects on the animals. They looked normal during the feeding trials and after. It is reported [Orlov, 1933] that the cake is extensively used for horse-feed. As the cake contains about 5 per cent nitrogen (N), 1.6 per cent phosphoric acid (P_2O_5) and 1.15 per cent potash (K_2O), it can be used as a good nitrogenous organic manure comparing favourably with an average sample of castor cake.

If facilities for the extraction or marketing of the oil do not exist, the seed can by itself be used as cattle food as it contains about 24 per cent crude proteins, 23 per cent albuminoids, 36 per cent ether extractives, 14 per cent carbohydrates, and is free from nicotine. As the seed has a thick seed-coat it should be soaked in water for two days, washed well, ground into a paste like horse-gram and fed to cattle.

Further studies on seed oil

Though the oil is fit for use for soap making and for illumination or edible purposes, its use as a semi-drying oil seems most advisable as the Madras province does not produce any other drying or semi-drying oil worth the name. For the supply of linseed oil which is the chief oil used for paint in the province, it depends entirely on northern India, and the typical drying oil - *tung* oil - is primarily imported from Burma and China.

The percentage composition of the different fatty acids in tobacco seed oil, linseed oil and *tung* oil are produced in Table VI, and some of their physical and chemical constants are given in Table VII. While *tung* oil stands unique in its composition and drying properties, linseed oil comes intermediate and tobacco-seed oil next in drying properties. The total absence of linolenic acid in tobacco-seed oil makes it dry somewhat slower, but its absence enables white paints mixed with tobacco-seed oil to continue to remain white, while they turn yellowish when mixed with linseed oil. While tobacco seed oil can be clarified to a practically colourless liquid, linseed oil cannot be so managed. These two characters of the oil make it eminently suited for the preparation and application of white paint and white enamels in preference to linseed oil.

TABLE II

*Some important physical and chemical constants of Gunter Virginia tobacco seed oil***

Analysed by	Specific gravity at 30°C.		Refractive index at 40°C.		Acid value		Saponification No.		Iodine No.		Butyro-refracto- meter at 40°C.		Nicotine	
	C. D.	H. D.	C. D.	H. D.	C. D.	H. D.	C. D.	H. D.	C. D.	H. D.	C. D.	H. D.	C. D.	H. D.
1. Indian Institute of Science, Bangalore	1.46850	1.46850	0.800	0.395	190.4	190.3	124.4	124.0	Absent	Absent
2. Imperial Agricultural Chemist, New Delhi	137.1	138.7
3. Government Food Analyst, Gulduy	0.55	0.39	142.0	142.0	64.7	65.3
4. Government Agricultural Chemist, Coimbatore	169.0	191.4	136.6	124.6
5. Superintendent, Kerala Soap Institute, Calicut	...	0.912	...	1.4725*	188.6	...	142.5
6. R. B. Technological Institute, Cawnpore	...	0.9153	...	1.4684	186.4	...	154.5

**C. D. = Cold drawn; H. D. = Hot drawn

* At 15.5°C.

† At 30°C.

TABLE III

Results of analysis (on dry basis) of Guntur Virginia tobacco seed cake for food and mineral values as against an average sample of gingelly cake

Particulars	Tobacco seed cake, Analysed by						Gingelly Analysis Govt. Chem Coimbatore
	Impl. Agrl. Chemist, New Delhi		Govt. Food Analyst, Guindy		Govt. Agrl. Chemist, Coimbatore		
	C. D.	H. D.	C. D.	H. D.	C. D.	H. D.	
<i>A. Food values</i> (per cent)							
Moisture	5·87	5·22
Ash	10·29	5·53	10
Crude proteins (N × 6·25)	30·75	35·81	30·58	34·13	37
Ether extrac- tives	16·00	17·08	17
Crude fibre	16·60	16·07	3
Carbohydrates (by difference).	26·53	27·19	30
Total	100·00	100·00	100
True proteins or albuminoids	28·52	33·81	35
Oil	11·2	14·5
Nicotine	Absent	Absent					
<i>B. Mineral values</i> (per cent)							
Lime (CaO)	On sample weight		
					0·65	0·22	
Potash (K ₂ O)	1·13	1·17	
Phosphoric acid (P ₂ O ₅)	2·5	2·6	1·85	1·41	
Nitrogen (N)	4·92	5·73	4·6	5·2	4·89	5·46	

C. D. = Cold drawn ; H. D. = Hot drawn

** Protein contents—

Gingelly cake from shelled seed	33·13	} G. A. C., Coimbatore
Gingelly cake from un-shelled seed	44·31	

TABLE IV

Chemical characters of the tobacco seed oil as determined by investigators in other parts of the world

Name of investigator	Ref. No.	Saponification value	Iodine value	Remarks
Strobel and Sourti [1904]	1	190.00	118.60	
Secker and Brezina [1919]	2	196.40	131.60	
W. [1920]	3	194.60	135.46	Used as edible and semi-drying oil
Samitskii [1929]	4	200.70	135.34	Mixed with driers and pigments; rather slow-drying films were obtained
Strobel and Grashin [1930]	5	190.49	139.74	
Strobel and Schutte [1934]	7	182.00	152.00	Nicotine absent
Strobel and Dedinszky [1934]	8	190.50	..	

TABLE V

Results of analysis of tobacco seed cake for the food values (on dry basis) as determined by investigators in other parts of the world

Particulars	Ref. 8	Ref. 10
	Per cent	Per cent
Moisture	3.38	8.66
Ash	4.97	14.43
Proteins	34.05	38.31
Fat	24.33	10.69
Fibre	23.64	17.97
Starch (digestible)	..	36.37
Odour	No tobacco odour	Has been successfully used for horse feeding; nicotine absent

TABLE VI

Percentage composition of different fatty acids in tobacco seed oil as compared those in linseed oil and tung oil

Name of acid	Tobacco seed oil			Linseed oil	Tung oil
	Ref. No. 4	Ref. No. 7	Ref. No. 11	(Ref. No. 12)	(Ref. No. 13)
	Per cent	Per cent	Per cent	Per cent	Per cent
Oleic.	21·70	16·20	26·37	5·00	10·00
Linoleic	60·00	70·40	60·23	48 to 59	..
Linolenic	21 to 32	..
Palmitic	9·60	3·10	7·03	} 10·00	..
Stearic	4·80	3·04		..
Elaeostearic	80·00
Glyceryl radicle	4·60	..
Unsaponifiable matter	1·20	1·25	1·41
Remarks	Alcoloids and linolenic acid absent

Economic aspect

At an estimate of 175 lb. seed per acre and 25 per cent of extractable oil in it, the province can supply about 8,800 tons of seed, or 2,200 tons of oil and 6,600 tons of cake per annum if the whole quantity of seed produced is gathered. If the oil is valued on par with the present price for linseed oil and cake on par with gingelly cake, the province can get annually enriched by 13 lakhs of rupees, if all the Virginia tobacco seed produced is utilized.* This will incidentally make the province, at least partly, self-sufficient in its requirements for semi-drying oils.

Several paint firms have placed bulk indents for the supply of oil. The quotations are, however, rather low, and the matter is under correspondence. A special grant of Rs. 500 has been sanctioned by the Madras Government to start business in this line.

*An acre of Virginia tobacco contains about 5,000 plants and seed from about 100 plants will be more than sufficient to produce seedlings to plant an acre.

TABLE VII

Important physical and chemical constants of Guntur Virginia tobacco seed oil, linseed oil and tung oil, as noted in their raw form

Particulars	Virginia tobacco seed oil (Guntur)	Linseed oil*	Tung oil*
Specific gravity at 15.5°C. . . .	0.912 to 0.915	0.931 to 0.941	0.939 to 0.943
Refractive index at 20°C. . . .	1.4684 to 1.4725	1.4742 to 1.4754	1.518 to 1.522
Acid number	0.39 to 0.80	Up to 10	Up to 5
Saponification number	186 to 191	189 to 196	189 to 195
Unsaponifiable matter	1.2 to 1.41**	0.8 to 2.0	Up to 0.75
Smoke value	124 to 155	170 to 185	155 to 167

Reference 13

Table IV

Further work

Bulk samples of the oil have been sent to several leading firms manufacturing enamels, paints and varnishes, throughout India and their reports awaited. Preliminary investigations made in the Sodhan Laboratories, Tenali, Guntur district, on the industrial utilization of the oil are sufficiently encouraging. Manurial trials with cake were arranged on all important crops grown under dry, irrigated and wet conditions during the year and they will be further elaborated during the next year. The technique of crushing is being studied for better extraction. If sufficiently engaging reports are received from the different manufacturing firms, colonization of seed and establishment of market for seed, oil and cake will be contemplated. In the meantime, necessary propaganda is being undertaken to utilize the seed as cattle food.

CONCLUSIONS

Virginia tobacco seed extensively produced in the Guntur, Kistna and Nellore districts contains nearly 35-37 per cent of edible and semi-drying

The oil can with advantage be used as semi-drying oil.
The cake can be fed to cattle or used as manure.

Till a market for seed, oil and cake is established, the seed can be sold to cattle.

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